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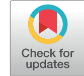
Obesity and Eating Disorders

Impaired Reverse Cholesterol Transport is Associated with Changes in Fatty Acid Profile in Children and Adolescents with Abdominal Obesity



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A B S T R A C T

Background: Abdominal obesity is an important cardiovascular disease risk factor. Plasma fatty acids display a complex network of both pro and antiatherogenic effects. High density lipoproteins (HDL) carry out the antiatherogenic pathway called reverse cholesterol transport (RCT), which involves cellular cholesterol efflux (CCE), and lecithin:cholesterol acyltransferase (LCAT) and cholesteryl ester transfer protein (CETP) activities.
Objectives: Our aim was to characterize RCT and its relation to fatty acids present in plasma in pediatric abdominal obesity.
Methods: Seventeen children and adolescents with abdominal obesity and 17 healthy controls were studied. Anthropometric parameters were registered. Glucose, insulin, lipid levels, CCE employing THP-1 cells, LCAT and CETP activities, plus fatty acids in apo B-depleted plasma were measured.
Results: The obese group showed a more atherogenic lipid profile, plus lower CCE (Mean±Standard Deviation) (6 ± 2 vs. 7 ± 2%; $P < 0.05$) and LCAT activity (11 ± 3 vs. 15 ± 5 umol/dL.h; $P < 0.05$). With respect to fatty acids, the obese group showed higher myristic (1.1 ± 0.3 vs. 0.7 ± 0.3; $P < 0.01$) and palmitic acids (21.5 ± 2.8 vs. 19.6 ± 1.9; $P < 0.05$) in addition to lower linoleic acid (26.4 ± 3.3 vs. 29.9 ± 2.6; $P < 0.01$). Arachidonic acid correlated with CCE ($r = 0.37$; $P < 0.05$), myristic acid with LCAT ($r = -0.37$; $P < 0.05$), palmitioleic acid with CCE ($r = -0.35$; $P < 0.05$), linoleic acid with CCE ($r = 0.37$; $P < 0.05$), lauric acid with LCAT ($r = 0.49$; $P < 0.05$), myristic acid with LCAT ($r = -0.37$; $P < 0.05$) ecoisatrienoic acid with CCE ($r = 0.40$; $P < 0.05$) and lignoseric acid with LCAT ($r = -0.5$; $P < 0.01$).
Conclusions: Children and adolescents with abdominal obesity presented impaired RCT, which was associated with modifications in proinflammatory fatty acids, such as palmitoleic and myristic, thus contributing to increased cardiovascular disease risk.

Keywords: HDL, abdominal obesity, cholesterol efflux, LCAT, CETP, fatty acid profile

Abbreviations: ALP, alkaline phosphatase; ALT, alanine aminotransferase; Apo, apolipoprotein; AST, aspartate aminotransferase; CCE, cellular cholesterol efflux; CETP, cholesteryl ester transfer protein; DBP, diastolic blood pressure; GGT, gamma-glutamyl transferase; HDL, high density lipoprotein; HOMA-IR, homeostasis model assessment insulin resistance; HsCRP, high sensitivity C-reactive protein; LCAT, lecithin:cholesterol acyl transferase; LDL, low density lipoprotein; Lp-PLA$_2$, lipoprotein-associated phospholipase A$_2$; QUICKI, quantitative insulin sensitivity check index; RCT, reverse cholesterol transport; SBP, systolic blood pressure; WC, waist circumference.

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Introduction

According to the World Health Organization (WHO), overweight and obesity are the sixth risk factor for human death worldwide. Every year, at least 3.4 million adults die as a result of being overweight or obese [1]. Furthermore, in 2016, more than 1.9 billion adults were overweight; in turn, 650 million were obese [1]. This means that 39% of adults are overweight, and 13% are obese worldwide. For children and adolescents, 39 million children under 5 y of age were overweight or obese in 2020 [1]. In turn, 340 million children and adolescents between the ages of 5 and 19 were overweight or obese in 2016 [1]. In the United States, according to the Centers for Disease Control, the prevalence of obesity in children and adolescents aged 2 to 19 y was 19.3% (14.4 million individuals) in the period between 2017 and 2018 [2].

Moreover, it is important to note that, in recent years, the fact that the distribution of adipose tissue is as important as the magnitude of fat deposition has become widely accepted. Therefore, the presence of specific abdominal obesity has been recognized as a major risk factor for cardiovascular disease (CVD) [3]. In this sense, abdominal obesity is specifically associated with the development of insulin resistance (IR) and type 2 diabetes mellitus (T2DM) in adults [4,5]. Furthermore, a series of studies carried out in adult populations have been published describing dyslipidemia in patients with central fat distribution. The syndrome is similar to the atherogenic dyslipidemia seen in association with general obesity [6,7]. In addition to IR and dyslipidemia, abdominal obesity is also closely associated with inflammation, both systemic and vascular specific in adult populations [8,9]. Regarding pediatric populations, few studies have shown a positive association between the presence of abdominal obesity, a higher risk of alterations in glucose metabolism, and presence of atherogenic dyslipidemia [10].

The presence of abdominal obesity has also been related to alterations in plasma fatty acid profile, which are associated with the development of both coronary artery disease and ischemic stroke [11,12]. Particularly, in a study carried out in elderly adults, abdominal obesity showed a positive correlation with palmitic acid concentration and a negative one with levels of linoleic acid [13]. Moreover, plasma fatty acids have been involved in the development of IR. In this regard, oleic acid has been reported to have a protective role against IR, whereas palmitic acid has the opposite effect [14,15]. Furthermore, fatty acid profile can also influence lipid levels. For example, stearic and linoleic acids are known to decrease low density lipoprotein cholesterol (LDL-C) concentration [16,17], whereas oleic acid can increase high density lipoprotein cholesterol (HDL-C) levels [18,19].

As previously explained, abdominal obesity is associated with the development of atherogenic dyslipidemia, mainly characterized by increased triglyceride and decreased HDL-C levels [6,7]. In this context, HDL constitutes the only lipoprotein fraction with cardioprotective properties. In fact, HDL particles possess multiple antiatherogenic functions, such as the promotion of reverse cholesterol transport (RCT), antioxidant activity, anti-inflammatory capacity, incorporation of free cholesterol from triglyceride-rich lipoproteins, and antiapoptotic function, among others [20,21]. RCT constitutes the most studied of these properties, which represents a complex process made up of different steps by which HDL particles transport excess cholesterol from peripheral tissues, including the arterial wall, to the liver for subsequent recycling or removal from the body [22]. The first step in RCT is free cholesterol efflux from peripheral tissue cells to HDL particles. In particular, HDLs remove cholesterol from macrophages, preventing lipid loading and foam cell formation, thus promoting atherosclerotic plaque regression [23]. The second and third steps of RCT are carried out by lecithin:cholesterol acyltransferase (LCAT) and cholesteryl ester transfer protein (CETP), respectively [24,25]. As far as we know, the status of RCT in children and adolescents with abdominal specific obesity has not been previously studied.

The aim of the present study was to analyze the different steps of RCT in children and adolescents with abdominal obesity and to explore a possible connection with the fatty acid profile.

Methods

Study design and subjects

A cross-sectional observational study was carried out. Seventeen children and adolescents of both sexes who presented abdominal obesity and 17 normal-weight controls were included. Abdominal obesity was defined as waist circumference (WC) $>$ 90% for the corresponding age and sex. All children and adolescents included in the study attended the Nutrition and Diabetes Service of the Children's Hospital “Dr. Ricardo Gutiérrez.” The study was approved by the Ethics Committee for Human Research of the Children Hospital “Dr. Ricardo Gutiérrez” and the Ethics Committee of the School of Pharmacy and Biochemistry of the University of Buenos Aires following the Helsinki Declaration and its later amendments or comparable ethical standards. Informed consent was obtained from all parents, and all the children signed the informed assent form. Inclusion criteria for controls were age between 8 and 17 y (shared with the obese group), BMI-z $<$ 1 and BMI $<$ 85 percentile according to WHO, and presence of normal lipid levels. Exclusion criteria for both the obese group and controls were the presence of any chronic or acute disease other than obesity, undergoing dietary or pharmacological treatment that could affect glucose and/or lipid levels or metabolism, and lack of a signed consent form. Regarding sociodemographic factors, all of the children and adolescents included in the study belonged to a similar socioeconomic background. They were all enrolled in public schools located in the vicinity of the Children's Hospital “Dr. Ricardo Gutiérrez” (within a 3-block radius) and inhabited middle-income households. Furthermore, even though they all possessed healthcare coverage, none of them had prepaid health insurance and, therefore, attended a public hospital such as the Children's Hospital “Dr. Ricardo Gutiérrez.”

Sample collection and processing

Blood samples were drawn from the antecubital vein after 12 h of fasting. A minimum of 10 mL of blood were drawn per participant. Aliquots were placed in tubes containing a coagulation accelerator or Na_2EDTA. Tubes were centrifuged at 1,500 x g for 15 min to obtain serum or plasma. Serum and plasma aliquots were stored at -70° C until their use.

Anthropometric parameters

Weight, height, WC, and Tanner stage were evaluated. WC was measured following the WHO STEPS protocol. The following

anthropometric indicators were calculated: BMI, BMI-z-score, and 90th percentile for WC. For these calculations, the WHO ANTHRO and ANTHRO Plus programs were used.

General biochemical determinations

Plasma glucose, hemoglobin, hematocrit, total protein, albumin, bilirubin, aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), gamma-glutamyl transpeptidase (GGT), urea and creatinine concentrations were determined by standardized methods (Roche, Basel, Switzerland). Plasma insulin concentration was measured with a microparticle enzyme immunoassay (MEIA, ABBOTT, Japan). HOMA-IR (Homeostasis Model Assessment) and QUICKI (Quantitative Sensitivity Check) indexes were calculated. Plasma high sensitivity C-reactive protein (hsCRP) was determined by a high sensitivity immunoturbidimetric assay (Roche, Basel, Switzerland).

Lipid profile

Plasma triglyceride (TG) and total cholesterol (TC) levels were determined by standardized methods (Roche, Basel, Switzerland). HDL fraction was isolated in the supernatant obtained after the selective precipitation of apo B-containing lipoproteins using phosphotungstic acid in the presence of magnesium ions. LDL-C levels were determined as the difference between TC and the cholesterol contained in the supernatant obtained after selective precipitation of LDL with polyvinyl sulfate in polyethylene glycol [26]. NonHDL-cholesterol (NonHDL-C) was calculated as the difference between TC and HDL-C. VLDL-C was obtained as the difference between the cholesterol of the supernatants of LDL-C and HDL-C determinations. Apo B and apo A-I were measured by immunoturbidimetry (Roche, Basel, Switzerland). Abnormal lipid levels were defined following the recommendations of the National Health Institute [27].

Lp-PLA$_2$ activity

Lp-PLA$_2$ activity was measured using a radiometric assay. Briefly, the incubation mixture contained 50 mL of 1/50 diluted serum and 10 mmol/L 1 hexadecyl-2-[3H]-acetyl-glycero-3-phosphocholine (specific activity 525 mCi.mmol/L) in a total volume of 0.5 mL of phosphate-buffered saline buffer (pH=7.4). Tritiated substrate 1-hexadecyl-2-[3H]-acetyl-glycero-3-phosphocholine (13.5 Ci.mmol/L) (New England Nucleotides) and nontritiated substrate (Cay-man Chemical) were mixed, the solvents were evaporated under nitrogen stream, redissolved in phosphate-buffered saline, and sonicated. Incubation was carried out at 37º C for 5 min, and the enzymatic reaction was stopped in an ice bath and with the addition of 1.5 mL of chloroform. Then, 0.5 mL of saturated sodium bicarbonate solution were added; after centrifugation, the aqueous phase was washed twice with 1.5 mL of chloroform. Radioactivity of the aqueous phase was measured by liquid scintillation using a Packard autoanalyzer. Radioactivity of the substrate buffer was also measured. Results were expressed as umol/mL.h. Measurements were all carried out in duplicate within the same assay. Within-run precision (CV) for Lp-PLA$_2$ activity was 5.1%

LCAT activity

LCAT activity was measured employing a radiometric method [28]. Briefly, the procedure utilized a substrate of heat-inactivated human plasma lipoproteins equilibrated with [^{3}H]-cholesterol plus albumin inactivated plasma (1:8:1 v/v). After heating human plasma at 57° C for 60 min, the substrate was recovered by centrifugation at 13,200 rpm for 3 min in an Eppendorf microfuge. Inactivated plasma was labeled with a stabilized cholesterol emulsion prepared by adding 1 nmol of [^{3}H]-cholesterol to one volume of 5% BSA in 150 Mm NaCl. Four volumes of inactivated plasma were added to the cholesterol emulsion and incubated at 37° C for 2–4 h. To start the enzyme activity assays, the substrate was mixed with one volume of enzyme in a final volume of 30 uL. After incubation at 37° C, the reaction was stopped with 200 uL of 1% digitonin (in 95% ethanol) and 10 uL of 5 mg/mL cholesterol. Samples were vigorously mixed and centrifuged at 6000 rpm for 10 min in an Eppendorf microfuge. Tritiated cholesteryl esters produced by LCAT were measured in 0.2 mL aliquots of the supernatant plus 2.5 mL of liquid scintillation cocktail. Results were expressed as umol/dL.h. Measurements were all carried out in duplicate within the same assay. Within-run precision (CV) for LCAT activity was 4.5%.

CETP activity

CETP activity was determined in serum samples following a radiometric method. Briefly, the ability of serum to promote the transfer of tritiated cholesteryl esters from the biosynthetically labeled HDL$_3$ fraction (^{3}H-CE-HDL$_3$) (NEN Life Science Products) to the apo B-containing lipoproteins present in the serum was evaluated. Serum samples were incubated with ^{3}H-CE-HDL$_3$ (50 umol/L cholesterol) and iodoacetate (1.5 mmol/L) in tris-buffered saline buffer (pH=7.4) for 3 h at 37° C. After incubation, apo B-containing lipoproteins were separated from HDL by the selective precipitation method using phosphotungstic acid (0.44 mmol/L) in the presence of magnesium ions. Radioactivity in the reaction mixture and in the supernatant containing the HDL fraction was measured in a liquid scintillation counter (Packard 210 TR; Packard Instruments, Meridians). Results were expressed as the percentage of cholesterol transported from HDL$_3$ particles to apo B-containing lipoproteins per mL per h. Measurements were all carried out in duplicate within the same assay. Within-run precision (CV) for CETP activity was 4.9%.

Cholesterol efflux determination

HDL was isolated by selective precipitation using polyethylene glycol (6000) (Sigma) in 10 mmol/L HEPES (pH=8.0) after centrifugation at 2200 x g and 4° C for 30 min. Apo B levels were measured by immunoturbidimetry in the supernatant to confirm the remotion of apo B-containing lipoproteins from plasma. The supernatant containing HDL was used for cellular cholesterol efflux (CCE) assay in THP-1 macrophages. Briefly, THP-1 cells were differentiated into macrophages by treatment with 100 nmol/L phorbol 12-myristate 13-acetate for 48 h. Subsequently, THP-1 macrophages were loaded with 1 μCi ^{3}H-cholesterol and 50 μg/mL acetylated LDL for 24 h. After that, 2% HDL-containing plasma was used as cholesterol acceptor for 4 h. Radioactivity was measured by liquid scintillation counting. CCE was calculated using the following equation: (radioactivity in efflux medium/total radioactivity in cells and medium) $\times$ 100. Results from efflux experiments carried out without acceptor were subtracted from each individual value. Results were expressed as the percentage of free cholesterol transported from cells to HDL particles. Measurements were all carried out in

duplicate within the same assay. Within-run precision (CV) for CCE determination was 4.7%.

Fatty acid profile

The fatty acid profile in apo B-depleted plasma was determined by gas chromatography (GC). Fatty acid methyl esters (FAMEs) from plasma were prepared according to a modified method of Lepage (CITA). Briefly, 2 mL of a methanol-toluene (4:1, v/v) were added to 200 μL of plasma, and then 0.2 mL of acetyl chloride were slowly added. After heating at 100° C for 1h, 5 mL of a 6% K_2CO_3 solution were added to the tube, mixed on a vortex, and centrifuged, and the clear toluene top layer containing FAMEs was obtained. FAMEs were analyzed using a Clarus 500 GC equipped with a Supelco SP 2560 100 m x 0.25 mm x 0.20 um column and flame ionization detector at 280 ° C, using helium as carrier gas. The plasma fatty acid data were expressed as the percentage (%) of total fatty acids (limit of quantification a value of 0.05%) [29,30].

Sample calculation and statistical analysis

The primary outcomes of the present study were CCE, LCAT activity, CETP activity (Figure 1), and fatty acid profile. Secondary outcomes were markers of glucose metabolism, basic lipid profile, liver enzymes, hsCRP, and Lp-PLA2. Sample calculation was performed employing Eppidata Software. Taking into account the main objectives of the present study, CCE and LCAT activity were employed as the selected variables, and the calculation was performed with unpublished data from a preliminary study carried out by our group (CCE: 7.78 ± 2.21 % for controls vs. 5.50 ± 1.96 % for obese children and adolescents; LCAT: 14.26 ± 2.90 umol/dL.h for controls vs. 10.67 ± 3.29 umol/dL.h for obese children and adolescents). The ratio of sample size was 1. The number of both patients and controls necessary to obtain 80% statistical power was estimated as 14 for CCE and 12 for LCAT activity. There was no missing data for any of the parameters. Variable distribution was assessed by the Shapiro Wilks test. Comparisons between populations were performed by Student t test for continuous normally distributed variables and Mann-Whitney U test for continuous nonnormally distributed ones. All variables analyzed with the Student T test were normally distributed according to the Shapiro Wilks test, and homogeneity of variance was assessed with the Levene test. In the case of variables analyzed with Mann-Whitney U test, nonnormality was established with Shapiro Wilks test, and histograms were employed to verify that the variables had similar distribution in the 2 groups. Analysis of covariance (ANCOVA) test was used to compare variables between groups, adjusting by sex, age, and Tanner stage. Only variables that remained statistically different between groups after correcting by sex, age, and Tanner stage were shown as such in tables and figures. In the ANCOVA models, normality of residuals was analyzed with the Shapiro Wilks test, and homogeneity of variance was assessed with the Levene test. Data were displayed as mean ± standard deviation or median (interquartile range) depending on data distribution. Multiple comparison problem was corrected by adjusting individuals' *P* values with the Benjamini-Hochberg Procedure. Pearson (parametric) and Spearman (nonparametric) correlation coefficients were employed to analyze univariate associations employing data from the whole population. Multiple testing problem was corrected by adjusting individuals *P* values with the Benjamini-Hochberg Procedure. Multiple Linear regression analyses were performed to explore the association between steps of RCT and individual fatty acids adjusting by the presence of obesity and the possible interaction between obesity and fatty acids. All variables were introduced in one step during the analysis. Normality of residuals was analyzed with the Shapiro Wilks test, and homogeneity of variance was assessed with the Levene test. All analyses were carried out with SPSS 19.0 statistical software.

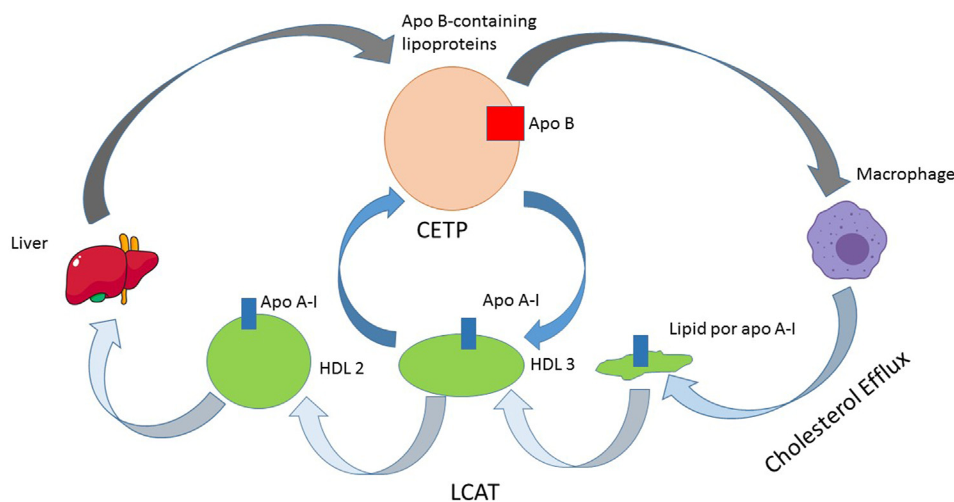

FIGURE 1. The 3 steps of RCT.
RCT represents the transfer of cholesterol from peripheral tissues to the liver. It involves different steps. The first step consists of free cholesterol transport through the cell membrane to HDL particles. The second step is catalyzed by LCAT, which esterifies cholesterol, increasing its hydrophobicity and promoting HDL maturation. The third step is carried out by CETP, which mediates the exchange of esterified cholesterol and TG between apo B-containing lipoproteins and HDL. RCT, reverse cholesterol transport; HDL, high density lipoprotein; LCAT, lecithin:cholesterol acyltransferase; CETP, cholesteryl ester transfer protein; TG, triglycerides; apo, apolipoprotein.

Results

The analysis of the antecedents and the anthropometric parameters of the studied population showed that the obese group had a higher frequency of first-degree relatives with a history of obesity, as well as higher values of z-BMI, WC, diastolic blood pressure (DBP-Z) and systolic blood pressure (SBP-Z) (Table 1). No significant differences were observed in the distribution of sexes, age, or Tanner stage (Table 1).

Table 2 shows the biomarkers of glucose metabolism, liver-related parameters, and hsCRP levels as a marker of systemic inflammation. The obese group showed higher plasma levels of insulin and nonesterified fatty acids and higher HOMA-IR plus lower QUICKI indexes, suggesting the presence of altered glucose metabolism. Furthermore, children and adolescents with abdominal obesity showed higher activity of ALT, FAL, and GGT and higher levels of hsCRP. In addition, the obese group also showed higher Lp-PLA_2 activity, considered a marker of vascular specific inflammation (Figure 2).

The analysis of the lipid parameters revealed that obese children and adolescents had higher levels of TG, VLDL-C, LDL-C, and nonHDL-C, increased TG/HDL-C, TC/HDL-C, and apo B/apo A-I ratios, in addition to lower levels of HDL-C and apo A-I. No significant differences were observed in TC and apo B concentrations (Table 3).

Regarding the fatty acid profile, children and adolescents with abdominal obesity showed higher levels of myristic, palmitic, and palmitoleic acids, together with lower concentrations of linoleic acid (Table 4).

Regarding RCT, the obese group showed significantly higher CCE (Figure 3, panel A) and LCAT activity (Figure 4). No statistically significant differences were observed in CCE/HDL-C (Figure 3, panel B), CCE/apo A-I (Figure 3, panel C), and CETP activity (Figure 5).

CCE was negatively associated with BMI-Z (r = -0.28; $P < 0.05$), ALP activity (r = -0.28; $P < 0.05$), and palmitoleic acid concentration (r = -0.38; $P < 0.05$), as well as positively related to arachidonic (r = 0.37; $P < 0.05$), linoleic (r = 0.37; $P < 0.05$) and ecoisatrienoic (r = 0.4; $P < 0.05$) acid levels, and LCAT activity (r = 0.40; $P < 0.05$). In turn, LCAT activity correlated positively with HDL-C (r = 0.43; $P < 0.01$) and apo A-I (r = 0.37; $P < 0.05$) levels and negatively with lauric (r = 0.49; $P < 0.05$), myristic (r = -0.37; $P < 0.05$) and lignoceric (r = -0.5; $P < 0.01$) acid concentrations. CETP activity was only associated with ALP activity (r = -0.32; $P < 0.05$). In linear regression analysis, the association between CCE and arachidonic acid (r^2 = 0.39; beta = 0.40; $P < 0.05$) remained statistically significant after adjusting for the presence of abdominal obesity and the interaction between abdominal obesity and arachidonic acid. The model met the normality of residuals and homogeneity of variance criteria.

In addition to the above-mentioned correlations, myristic acid correlated with BMI-z, WC, HOMA-IR, nonesterified fatty acids, and HDL-C. Palmitic acid correlated with BMI-z, HOMA-IR, and HDL-C. Palmitoleic acid correlated with BMI-z, WC, insulin, HOMA-IR, QUCKI, nonesterified fatty acids, ALT, AST, ALP, GGT, hsCRP, HDL-C, and apo A-I. Linoleic acid correlated with BMI-z, WC, nonesterified fatty acids, HDL-C, and apo A-I. Alpha-linoleic acid correlated with TG. Eicosapentaenoic acid correlated with nonesterified fatty acids, AST, ALT, and HDL-C. Finally, lignoceric acid correlated with nonesterified fatty acids (Table 5).

TABLE 1
Anthropometric parameters in children and adolescents with abdominal obesity and in controls with normal weight

Parameter	Controls with normal weight (n = 17)	Patients with abdominal obesity (n = 17)
Age (y)	13.1 ± 2.0	12.4 ± 2.7
Sex (F/M)	8 / 9	10 / 7
BMI-Z	0.5 ± 0.8	3.2 ± 0.8*
TannerStage I/Total	4 / 17	6 / 17
WC (cm)	70.8 ± 9.2	101.3 ± 13.7*
Incidence of abdominal obesity in first grade relatives (%)	18	59*
DBP-Z	-0.07 ± 0.01	0.65 ± 0.10 *
SBP-Z	-0.27 ± 0.03	1.54 ± 0.14 *

F, female; M, male; BMI-Z, BMI Z-score; WC, waist circumference; DBP-Z, diastolic blood pressure Z-score; SBP-Z, systolic blood pressure Z-score.
* $P < 0.05$ vs. controls with normal weight. Categorical variables were analyzed by Pearson chi-square. Measures shown as mean ± standard deviation for normally distributed variables. Parameters were compared by Student t test for independent samples.

TABLE 2
Biochemical parameters in children and adolescents with abdominal obesity and in controls with normal weight.

Parameter	Controls with Normal Weight (n = 17)	Patients with Abdominal Obesity (n = 17)
Glucose (mmol/L)	4.83 (4.44 – 4.89)	4.78 (4.61 – 5.06)
Insulin (pmol/L)	62.5 (48.6 – 76.4)	90.3 (76.4 – 159.7)*
HOMA-IR	1.7 (1.0–1.9)	2.7 (2.3–4.9)*
QUICKI	0.36 (0.34–0.40)	0.32 (0.30 – 0.33)*
Nonesterified fatty acids (mmol/L)	0.3 (0.2 – 0.5)	0.6 ± (0.5 – 0.8)*
Total protein (g/L)	76 (75 – 79)	76 (71 – 78)
Albumin (mmol/L)	0.71 (0.69 – 0.74)	0.71 (0.70 – 0.74)
Total Bilirubin (mmol/L)	0.007 (0.005 – 0.007)	0.009 (0.005 – 0.012)
AST (μkat/L)	0.33 ± 0.08	0.38 ± 0.13
ALT (μkat/L)	0.18 (0.15 – 0.30)	0.28 (0.22 – 0.33)*
ALP (μkat/L)	2.6 ± 1.4	3.6 ± 1.4*
GGT (μkat/L)	0.18 ± 0.07	0.31 ± 0.13*
HsCRP (nmol/L)	13.3 ± 9.5	28.6 ± 23.8*

AST, aspartate aminotransferase; ALT, alanine aminotransferase; ALP, alkaline phosphatase; GGT, gamma-glutamyl transpeptidase; hsCRP, high sensitivity C-reactive protein.
* $P < 0.05$ vs. controls with normal weight. Measures are shown as mean ± standard deviation for normally distributed variables, which were compared by Student t test for independent samples. Measures for nonnormally distributed variables are shown as median (interquartile range) and were compared with Mann-Whitney U test. ANCOVA test was performed to verify that differences remained statistically significant after adjusting by sex, age, and Tanner Stage. Multiple testing problem was corrected by adjusting individuals P values with the Benjamini-Hochberg Procedure.

Discussion

Children and adolescents with abdominal obesity presented higher incidence of abdominal obesity in first grade relatives, as

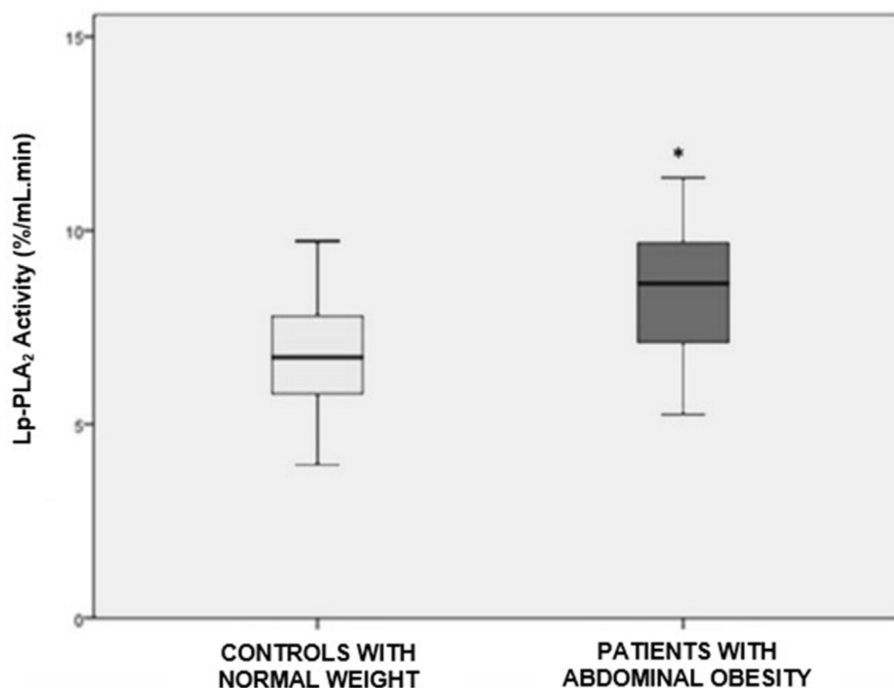

FIGURE 2. Lp-PLA$_2$ activity in children and adolescents with abdominal obesity and in normal weight controls.
Box-plot showing the comparison of Lp-PLA$_2$ activity between the groups. Lp-PLA$_2$, lipoprotein-associated phospholipase A$_2$. *$P < 0.05$ vs. normal weight controls according to Mann-Whitney U test. ANCOVA test was performed to verify that differences remained statistically significant after adjusting by sex, age, and Tanner stage.

well as higher values of BMI-Z, WC, TAD-Z, and TAS-Z. No significant differences were observed in the distribution of sexes, age, or Tanner stage. In turn, the obese group showed higher plasma levels of insulin and nonesterified fatty acids, higher HOMA-IR index, and lower QUICKI index, indicating the presence of alterations in glucose metabolism. Furthermore, children and adolescents with abdominal obesity showed higher activities

TABLE 3
Lipid and lipoprotein profile in children and adolescents with abdominal obesity and in controls with normal weight

Parameter	Controls with normal weight ($n = 17$)	Patients with abdominal Obesity ($n = 17$)
TG (mmol/L)	0.76 (0.55–1.03)	1.12 (1.02–1.37)*
TC (mmol/L)	3.95 ± 0.96	4.24 ± 0.67
LDL-C (mmol/L)	2.09 ± 0.88	2.66 ± 0.72*
HDL-C (mmol/L)	1.52 ± 0.34	1.18 ± 0.21*
Non-HDL-C (mmol/L)	2.48 ± 0.88	3.23 ± 0.75*
VLDL-C (mmol/L)	0.39 ± 0.16	0.57 ± 0.26*
Apo B (mmol/L)	26.42 ± 6.78	84 ± 20
Apo A-I (mmol/L)	50.71 ± 9.29	43.57 ± 5.71*
TG/HDL-C	1.2 (1.1–1.6)	2.6 (2.1–3.9)*
TC/HDL-C	2.7 ± 0.6	4.6 ± 1.6*
Apo B/apo A-I	0.4 ± 0.1	0.7 ± 0.3*

TG, triglycerides; TC, total cholesterol; LDL-C, low density lipoprotein cholesterol; HDL-C, high density lipoprotein cholesterol; VLDL-C, VLDL cholesterol; Apo, apolipoprotein.
* $P < 0.05$ vs. controls with normal weight. Measures are shown as mean ± standard deviation for normally distributed variables, which were compared by Student t test for independent samples. Measures for non-normally distributed variables are shown as median (interquartile range) and were compared with Mann-Whitney U test. ANCOVA test was performed to verify that differences remained statistically significant after adjusting by sex, age, and Tanner Stage. Multiple testing problem was corrected by adjusting individuals *P* values with the Benjamini-Hochberg Procedure.

TABLE 4
Fatty acid profile in apo B-depleted plasma from children and adolescents with abdominal obesity and controls with normal weight

Parameter	Controls with Normal Weight ($n = 17$)	Patients with Abdominal Obesity ($n = 17$)
Lauric acid	0.10 (0.08–0.14)	0.12 (0.09–0.20)
Myristic acid	0.72 ± 0.25	1.12 ± 0.29*
Palmitic acid	19.57 ± 1.98	21.51 ± 2.83*
Palmitoleic acid	1.85 ± 0.31	2.63 ± 0.43*
Margaric acid	0.54 ± 0.13	0.65 ± 0.18
Stearic acid	8.68 ± 0.92	8.90 ± 1.50
Oleic acid	17.62 ± 1.37	17.66 ± 1.70
Linoleic acid	29.93 ± 2.57	26.36 ± 3.31*
Gadoleic acid	0.14 ± 0.03	0.12 ± 0.03
Alpha-linoleic acid	0.21 (0.17–0.28)	0.25 (0.20–0.33)
Ecoisatrienoic acid	0.74 ± 0.15	0.67 ± 0.14
Arachidonic acid	4.64 (3.80–5.56)	4.86 (4.07 6.84)
Eicosapentaenoic acid	0.32 (0.19–0.70)	0.22 (0.18–0.27)
Lignoceric acid	0.35 ± 0.24	0.41 ± 0.12
Docosahexaenoic acid	0.68 (0.54–0.89)	0.74 (0.55–1.04)

Results expressed as area %. Apo B, apolpoprotein B.
* $P < 0.05$ *vs.* controls with normal weight. Measures are shown as mean ± standard deviation for normally distributed variables, which were compared by Student t test for independent samples. Measures for nonnormally distributed variables are shown as median (interquartile range) and were compared with the Mann-Whitney U test. ANCOVA test was performed to verify that differences remained statistically significant after adjusting by sex, age, and Tanner stage. Multiple testing problem was corrected by adjusting individuals *P* values with the Benjamini-Hochberg Procedure.

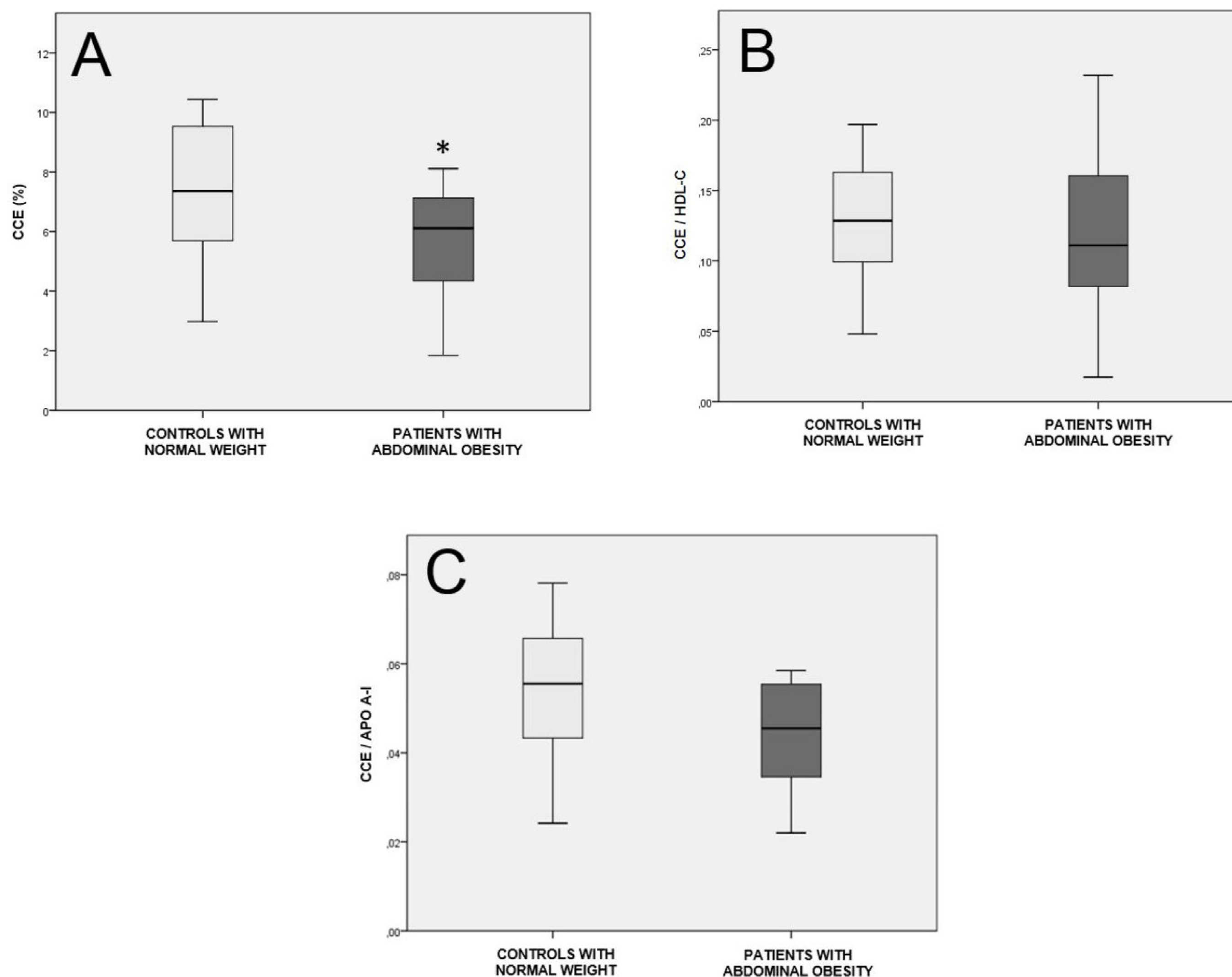

FIGURE 3. CCE in children and adolescents with abdominal obesity and in normal weight controls.
Box-plot showing the comparison of CCE (Panel A), CCE/HDL-C (Panel B), and CCE/apo A-I (Panel C) between the groups. CCE, cellular cholesterol efflux; HDL-C, high density lipoprotein cholesterol; apo A-I, apolipoprotein A-I. * $P < 0.05$ vs. normal weight controls according to Mann-Whitney U test. ANCOVA test was performed to verify that differences remained statistically significant after adjusting by sex, age, and Tanner stage.

of ALT, FA, L, and GGT, plus higher levels of hsCRP and Lp-PLA_2 activity. The analysis of lipid parameters revealed that the obese group had higher TG, VLDL-C, LDL-C, and nonHDL-C levels, as well as increased TG/HDL-C, TC/HDL-C, and apo B/apo A-I ratios, in addition to lower levels of HDL-C and apo A-I. Regarding RCT, the obese group exhibited lower CCE and LCAT activity, with no differences in CETP activity.

In our study, the group with abdominal obesity presented altered glucose metabolism, which coincides with previous results from our group [31] and from other authors [32]. Furthermore, the Bogalusa Heart Study reported an association between WC values and insulin levels in children and adolescents [33]. In fact, previous studies have also shown an association between abdominal fat deposits, measured by magnetic resonance imaging, and HOMA-IR values in children and adolescents [34]. In this regard, abdominal fat deposits were also associated with HOMA-IR in adolescents when quantified by ultrasound [35]. In turn, in another study, an association was reported between increases in visceral adiposity and decreased insulin sensitivity [36]. Consistently, Galcheva et al. reported higher levels of insulin and HOMA-IR in prepubertal children with abdominal obesity [37]. Furthermore, it should be noted that in a previous study carried out in Argentina, the HOMA-IR and the TG/HDL-C indexes were associated with CVD in children aged between 6 and 9 y [38]. To the best of our knowledge, our study would be the first to analyze glucose metabolism in a pediatric population with abdominal obesity, which included both children and adolescents. In addition to significant differences in traditional parameters of glucose metabolism, the obese group also presented increased levels of nonesterified fatty acids. This parameter is associated with obesity and serves as a marker of the presence of IR, which increases its release from visceral adipose tissue [39]. In summary, the combination of higher levels of insulin and nonesterified fatty acids, together with

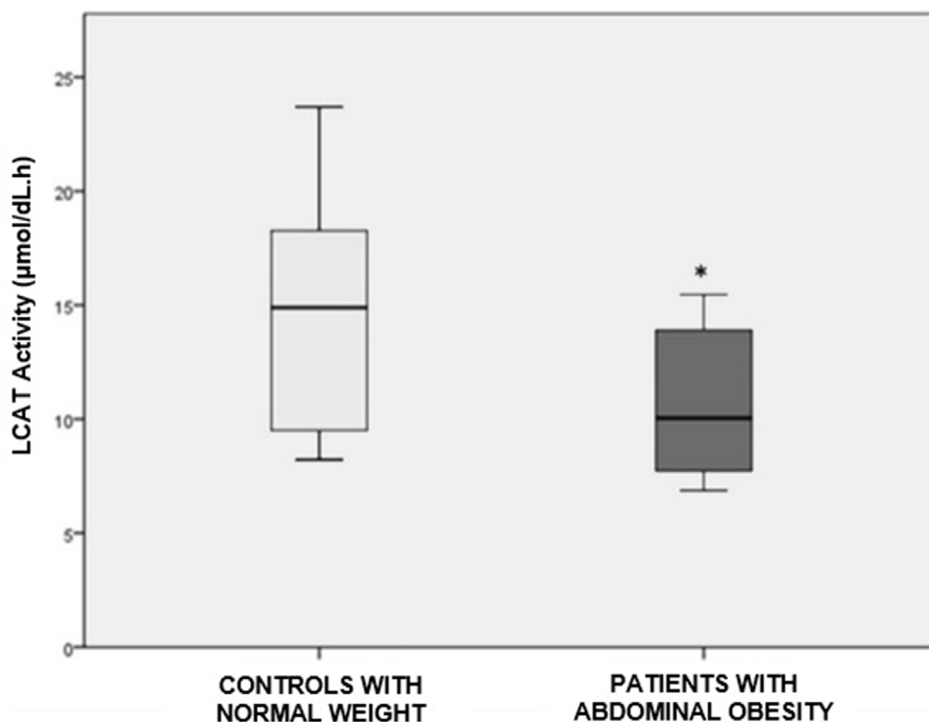

FIGURE 4. LCAT activity in children and adolescents with abdominal obesity and in normal weight controls.
Box-plot showing the comparison of LCAT activity between the groups. LCAT, lecithin:cholesterol acyltransferase. * $P < 0.05$ vs. normal weight controls according to Mann-Whitney U test. ANCOVA test was performed to verify that differences remained statistically significant after adjusting by sex, age, and Tanner stage.

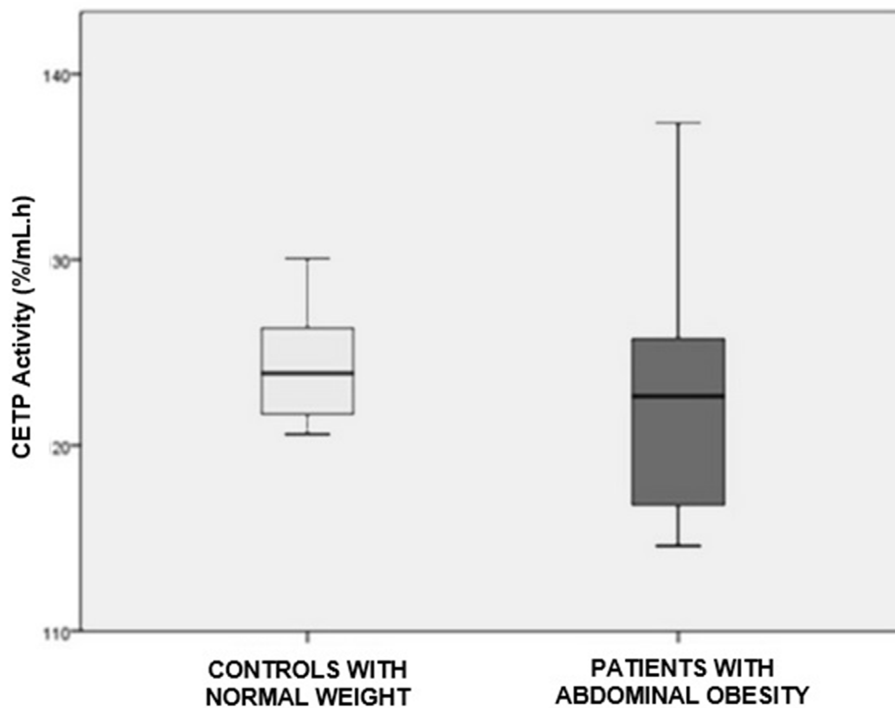

FIGURE 5. CETP activity in children and adolescents with abdominal obesity and in normal weight controls.
Box-plot showing the comparison of CETP activity between the groups. CETP, cholesteryl ester transfer protein. Comparison was carried out with Mann-Whitney U test.

TABLE 5
Correlations between fatty acid levels and general biochemical parameters

Parameter	BMI-Z	WC	Insulin	HOMA-IR	QUICKI	NEFA	ALT	AST	ALP	GGT	HsCRP	TG	HDL-C	Apo A-I
Myristic acid	0.48**	0.49**	0.35	0.40*	-0.26	0.38*	0.24	0.24	0.36	0.27	0.17	0.15	-0.37*	-0.21
Palmitic acid	0.35*	0.29	0.32	0.40*	-0.28	0.35	0.24	0.11	0.20	0.14	0.19	0.09	-0.34*	-0.13
Palmitoleic acid	0.73**	0.65**	0.39*	0.45**	-0.35*	0.64**	0.60**	0.38*	0.59**	0.53**	0.51**	0.18	-0.35*	-0.42*
Linoleic acid	-0.50**	-0.51**	-0.06	-0.16	0.02	-0.47*	-0.23	-0.18	-0.11	-0.27	-0.09	-0.11	0.38*	0.38*
Alpha-linoleic acid	0.16	0.29	0.36*	0.16	-0.24	0.18	0.21	0.19	0.31	0.14	-0.26	0.51**	-0.14	-0.17
Eicosapentaenoic acid	-0.37	-0.17	-0.14	-0.16	0.12	-0.49*	-0.40*	-0.39*	-0.41*	-0.21	-0.31	-0.18	0.40*	0.24
Lignoceric acid	0.26	0.27	0.12	0.13	-0.03	0.47*	0.22	0.20	0.03	0.09	0.20	-0.13	-0.14	-0.06

WC, waist circumference; HOMA-IR, Homeostatic Model Assessment for Insulin Resistance; QUICKI, Quantitative Insulin Sensitivity Check Index; NEFA, nonesterified fatty acids; ALT, alanine aminotransferase; AST, aspartate aminotransferase; ALP, alkaline phosphatase; GGT, gamma-glutamyl transferase; HsCRP, high sensitivity C-reactive protein; HDL-C, high density lipoprotein cholesterol; Apo, apolipoprotein.
*$P < 0.05$
** $P < 0.01$. Correlations were analyzed with Spearman coefficient. Multiple testing problem was corrected by adjusting individuals P values with the Benjamini-Hochberg Procedure.

higher HOMA-IR and TG/HDL-C and lower QUICKI indexes, could be indicative of the presence of alterations in glucose metabolism in this population, even at an early age.

The obese group also showed alterations in parameters traditionally associated with liver status, ALT, FAL, and GGT, suggesting the presence of low-grade liver inflammation. In this regard, a previous study showed a positive association between WC values and GGT activity in children and adolescents [40]. Furthermore, in another study, an association was reported between visceral adipose tissue levels, the presence of hepatic steatosis, and plasma ALT and GGT values in children and adolescents [41]. Accordingly, in other works, abdominal obesity was reported as a predictor of the presence of nonalcoholic fatty liver in preschool children [42] and adolescents [43]. Again, our study would be the first to analyze these markers of liver damage in children and adolescents who specifically presented abdominal obesity.

The group with abdominal obesity exhibited significantly higher levels of hsCRP than controls, indicating the presence of systemic inflammation. A previous study showed an association between the levels of hsCRP and WC in prepubertal children with abdominal obesity [37]. Other studies have reported this same association in children and adolescents with overweight and obesity [44,45]. In the present study, the obese group also showed higher Lp-PLA_2 activity. Lp-PLA_2 is considered a marker of specific vascular inflammation and a novel risk factor for cardiovascular disease [46,47]. Lp-PLA_2 is an enzyme secreted by inflammatory cells mainly associated with LDL particles in circulation [48]. It carries out the hydrolysis of oxidized phospholipids, liberating proatherogenic and proinflammatory oxidized fatty acids and lysophosphatidylcholine into the subendothelium [48,49]. This finding suggests the presence of vascular specific inflammation, which is associated with increased CVD risk, in addition to the presence of systemic inflammation [50,51]. A prior study reported higher Lp-PLA_2 activity in prepubertal children with obstructive sleep apnea and general obesity (50). Furthermore, in a previous study, our group found increased Lp-PLA_2 activity in children and adolescents with general obesity [51]. The present study would be the first to evaluate the activity of Lp-PLA_2 in children and adolescents with abdominal obesity.

Regarding the lipid profile, the obese group showed higher levels of TG, VLDL-C, LDL-C, and nonHDL-C, increased TG/HDL-C, TC/HDL-C, and apo B/apo A-I, in addition to lower levels of HDL-C and apo A-I. The presence of a more atherogenic lipoprotein profile is consistent with data reported in the literature on the subject. In a previous study, an association was reported between the presence of abdominal obesity and dyslipidemia in school-age children [42]. In fact, in another study, an increase in TG and TC levels was observed in prepubertal children with abdominal obesity [37]. Furthermore, another study reported an association between the presence of abdominal obesity and high levels of TG and low levels of HDL-C in prepubertal children [36]. The observed alterations, characterized by higher TG and lower HDL-C and apo A-I, are consistent with the presence of obesity-induced atherogenic dyslipidemia [52]. The higher levels of TG and lower HDL-C concentration found in the study are also consistent with the presence of altered glucose metabolism described above and possibly IR. In fact, IR causes an increase in the release of nonesterified fatty acids from adipose tissue. In the presence of this increased nonesterified fatty acid

flux, intrahepatic TG production is promoted, with consequent augmentation of hepatic synthesis and secretion of triglyceride-enriched VLDL into the bloodstream [53]. The expanded pool of TG-enriched VLDL and the increased activity of CETP produce an increased bidirectional exchange between TG of VLDL and cholesteryl esters of HDL particles [53].

It is important to note that recent studies have challenged the relevance of HDL-C as a marker of CVD risk [54]. In fact, different experts have recently suggested that the evaluation of HDL functionality should replace the measurement of other HDL-related parameters in terms of obtaining a more accurate estimation of atherogenic risk [55]. HDL capacity to promote CCE would be particularly important and partially independent of HDL-C and apo A-I concentration [56]. In our study, the group with abdominal obesity presented HDL particles with less capacity to promote CCE from THP-1 macrophages. In a previous study, a negative association between WC values and CCE was observed in children 5 to 9 y old who presented abdominal obesity [57]. Furthermore, in another study, 6 microRNAs synthesized by visceral adipocytes were identified, which would have a negative regulatory effect on CCE in adolescents [58]. In the present study, we found no statistically significant differences in CCE/HDL-C and CCE/apo A-I ratios. The lack of differences in these 2 ratios suggests that the lower CCE observed in the obese group could be the result of a lower number of HDL particles in circulation. In addition to the decrease in CCE, the group with abdominal obesity also had lower LCAT activity. LCAT activity is considered the driving force of RCT [24]. Information regarding LCAT activity in populations with abdominal obesity is scarce. In a recent work, LCAT activity was reported to be negatively associated with visceral fat accumulation in adults [59]. Our study would be the first to analyze CCE and LCAT activity in a pediatric population with abdominal obesity compared with healthy controls. In a prior study, we reported impaired CCE and LCAT activity in children and adolescents with general obesity [51]. The fact that the obese group presented lower CCE in conjunction with lower LCAT activity suggests a possible defect in HDL maturation and impairment of ABCG-1- mediated CCE. Finally, in the present study, we found no significant differences in CETP activity between the groups, which appears inconsistent with the increase in TG levels observed in obese patients. However, it should be noted that even though the obese group had higher TG, this parameter was within the acceptable range in most children and adolescents, showing only a minor increment. The joint presence of decreased CCE and LCAT activity in these children and adolescents suggests the existence of alterations in RCT and a deterioration in HDL functionality.

In our study, children and adolescents with abdominal obesity displayed higher levels of myristic, palmitic, and palmitoleic acids, as well as lower concentrations of linoleic acid. A previous work reported higher palmitic and palmitoleic acid levels in obese children; however, this study did not include adolescents, and the children presented general obesity [60]. Moreover, studies have shown higher palmitoleic acid in children with general obesity, with one of them identifying waist-to-hip ratio (WHR) as an independent predictor of its levels [61,62]. In concordance, a recent article found higher palmitoleic acid in obese children and adolescents [63]. Nevertheless, in the mentioned study, authors analyzed fatty acid profile in whole plasma, and obesity was defined according to BMI and WHR, whereas in our study, we employed WC percentile to define abdominal obesity, as recommended by WHO. In another study, authors reported higher levels of palmitoleic acid in children with abdominal obesity but did not include adolescents as part of the study population [64]. A possible explanation for the association observed between the presence of obesity and higher levels of palmitoleic acid would be Δ9 desaturase activity. This enzyme catalyzes the conversion of palmitic to palmitoleic acid, a process highly dependent on the plasma levels of insulin and glucose [65,66]. In fact, Δ9 desaturase activity has been proposed as a predictor of cardiometabolic risk [65]. Similarly, the higher levels of palmitic and myristic acids, together with lower concentration of linoleic acids, could explain, at least in part, the alterations in markers of glucose metabolism and inflammation observed in the obese group. Indeed, palmitic acid is known to promote inflammation and IR [67]. Furthermore, myristic acid could be associated with inflammation and IR in obese adolescents [68]. In fact, its supplementation increases inflammation in animal models [69]. On the contrary, linoleic acid, an omega-6 polyunsaturated fatty acid, would have beneficial effects on metabolism. In this regard, evidence suggests linoleic acid could present an anti-inflammatory effect [70,71]. Nevertheless, high levels of oxidized derivates of linoleic acid are associated with metabolic syndrome in children [72]. It should be noted that omega-6 fatty acids are generally considered proinflammatory, and the interaction of these fatty acids with their omega-3 counterparts, traditionally considered anti-inflammatory, is complex and not fully understood [70]. It is worth mentioning that our study would be the first to analyze fatty acid profile in apo B-depleted plasma, whereas prior studies have employed whole plasma for its determination. This fact could explain the differences observed between our findings and those reported in the bibliography.

Regarding associations between RCT and fatty acids, we found that CCE was negatively associated with palmitoleic acid and positively associated with arachidonic, linoleic, and ecoisatrienoic acids. Furthermore, LCAT activity showed negative association with lauric, myristic, and lignoceric acids. Arachidonic acid is an omega-6 polyunsaturated fatty acid, and its supplementation has beneficial effects on LDL-C and HDL-C levels, reducing risk of CVD (73). Moreover, bioactive leukotrienes produced by arachidonic acid metabolism could promote RCT [73]. However, evidence also suggests a negative effect of arachidonic acid on ABCA1 levels, a membrane transporter of crucial importance for CCE [74]. Furthermore, leukotrienes synthesized from arachidonic acid by 5-lipoxygenase display potent inflammatory effects [75]. As previously mentioned, linoleic acid has a protective effect against IR and inflammation [70,71]. Evidence regarding its effect on RCT appears contradictory. On one hand, linoleic acid supplementation suppressed ABCA1 and ABCG1 expression in murine macrophages. To the contrary, an oxidized derivative, such as 13-hydroxy linoleic acid, promoted cholesterol efflux in vitro [76,77]. In turn, ecoisatrienoic acid is an omega-9 polyunsaturated acid with anti-inflammatory properties [78]. Myristic acid is a saturated fatty acid that could promote IR and inflammation [68]. Regarding its effect on LCAT activity, evidence is also contradictory. In a prior study, a diet rich in myristic acid was shown to reduce LCAT activity [79]. On the contrary, moderate intake could increase LCAT activity [80]. Lauric acid is another saturated fatty acid, and longitudinal studies have shown an association between its consumption and

risk of CVD [81]. In fact, lauric, palmitic, and myristic acids would have a potent hypercholesterolemic effect [82]. Lignoceric acid is a very-long-chain saturated fatty acid. Studies have reported contradictory evidence regarding its association with CVD [83–85]. Palmitoleic acid is of particular interest. Previous studies have shown increases in the levels of this fatty acid in conditions such as abdominal obesity [64]. Such an increase would be associated with higher metabolic risk [63]. In fact, palmitoleic, as well as myristic and oleic acids, could be directly associated with risk of type 2 diabetes mellitus independently of insulin sensitivity [86]. Moreover, a recent study performed in murine models found that palmitoleic acid could contribute to vascular complications in diabetes [87]. Nevertheless, other studies have reported that palmitoleic acid could have an anti-inflammatory effect and improve insulin sensitivity [88, 89]. In our study, palmitoleic acid showed positive associations with BMI-Z, WC, insulin, HOMA-IR, nonesterified fatty acids, ALT, AST, ALP, GGT, and hsCRP, as well as negative correlations with QUICKI index, HDL-C, apo A-I, and CCE. These findings suggest that palmitoleic acid could play a role in the intricate network connecting inflammation, glucose metabolism, and lipid alterations in abdominal obesity. As far as we know, our study would be the first to report associations between fatty acids and different steps of RCT in obese children. Furthermore, the association between CCE and arachidonic acid remained statistically significant after adjusting for the presence of abdominal obesity and the interaction between the latter and arachidonic acid levels. This finding suggesting that individual fatty acids, such as arachidonic acid, could have specific effects on RCT that are independent of the presence of obesity. Fatty acids could have differential effects on separate steps of RCT enhancing or ameliorating HDL functionality and, therefore, represent a target of particular interest in the study of atherogenic risk evolution since the early stages of life.

In summary, children and adolescents with abdominal obesity showed alterations in glucose and lipid metabolism, as well as both systemic and vascular specific inflammation. Moreover, this population also showed impaired CCE and LCAT activity. These alterations were associated in diverse ways with fatty acid profile characterized in apo B-depleted plasma. These findings highlight the importance of the interaction between metabolic pathways in the progression of obesity and related complications.

Different limitations must be acknowledged. First, the study design was cross-sectional, and causality cannot be implied. Second, even though the sample size was enough to achieve 80% power, it was still small, and the results should be carefully interpreted. Third, glucose metabolism and liver status were assayed with parameters such as HOMA-IR, QUICKI, and liver enzymes, which present multiple limitations. Other assays such as oral glucose tolerance test, hyperinsulinemic-euglycemic clamp, or FIBROMAX panel would be useful in future studies. Fourth, due to the CCE assay employed, it is not possible to fully determine which membrane transporter could be involved in the CCE impairment observed in the obese group.

Author Contributions

The authors' responsibilities were as follows—Martin, M: Conceptualization, Design, Methodology, Investigation, Writing, Final content; Impa Condori, A: Conceptualization, Design, Methodology, Investigation, Writing, Final content; Davico, B: Conceptualization, Methodology, Investigation, Writing, Final content; Gómez Rosso, L: Writing- Reviewing and Editing, Final content; Gaete, L: Conceptualization, Methodology, Investigation, Writing, Final content; Tetzlaff, W F: Investigation, Writing, Final content; Lozano Chiappe, E: Investigation, Writing, Final content; Sáez, MS: Investigation, Writing, Final content; Lorenzon González, MV: Investigation, Writing, Final content; Godoy, MF: Investigation, Writing, Final content; Osta, V: Investigation, Writing, Final content; Trifone, L: Conceptualization, Methodology, Investigation, Writing, Final content; Ballerini, MG: Writing- Reviewing and Editing, Final content; Cherñavsky, A: Investigation, Writing, Final content; Boero, L: Writing- Reviewing and Editing, Final content; Tonietti, M: Conceptualization, Design, Methodology, Writing- Reviewing and Editing, Final content; Feliu S: Conceptualization, Design, Methodology, Writing- Reviewing and Editing, Final content; Brites, F: Conceptualization, Design, Methodology, Writing-Reviewing and Editing, Final content.

All authors have read and approved the manuscript.

MM, ICA, DB, GRL, GL, TWF, LCE, SMS, LGMV, GMF, OV, TL, BMG, CA, BL, TM, FS, and BF have read and approved the final version.

Conflict of interests

The authors declare that they have no conflict of interest.

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Data Availability

The datasets generated during and/or analyzed during the current study are available from the corresponding author upon reasonable request.

References

[1] https://www.who.int/news-room/fact-sheets/detail/obesity-and-overweight. Last accessed, 16/May/2023.
[2] https://www.cdc.gov/obesity/data/childhood.html. Last accessed 16/May/2023.
[3] C. Zhang, K.M. Rexrode, R.M. van Dam, T.Y. Li, F.B. Hu, Abdominal obesity and the risk of all-cause, cardiovascular, and cancer mortality: sixteen years of follow-up in US women, Circulation 117 (2008) 1658–1667, https://doi.org/10.1161/CIRCULATIONAHA.107.739714.
[4] S. Yaturu, Obesity and type 2 diabetes, J. Diabetes Mellitus. 4 (2011), https://doi.org/10.4236/jdm.2011.14012.
[5] S.R. Kashyap, R.A. Defronzo, The insulin resistance syndrome: physiological considerations, Diab. Vasc.. Dis. Res. 4 (2007) 13–19, https://doi.org/10.3132/dvdr.2007.001.
[6] J.P. Després, M. Ferland, S. Moorjani, A. Nadeau, A. Tremblay, P.J. Lupien, et al., Role of hepatic-triglyceride lipase activity in the association between intra-abdominal fat and plasma HDL cholesterol in obese women, Arteriosclerosis 9 (1989) 485–492, https://doi.org/10.1161/01.atv.9.4.485.

[7] J.P. Després, S. Moorjani, M. Ferland, A. Tremblay, P.J. Lupien, A. Nadeau, et al., Adipose tissue distribution and plasma lipoprotein levels in obese women. Importance of intra-abdominal fat, Arteriosclerosis 9 (1989) 203–210, https://doi.org/10.1161/01.atv.9.2.203.

[8] M.S. Ellulu, I. Patimah, H. Khaza'ai, A. Rahmat, Y. Abed, Obesity and inflammation: the linking mechanism and the complications, Arch. Med. Sci. 13 (2017) 851–863, https://doi.org/10.5114/aoms.2016.58928.

[9] J.P. Després, Inflammation and cardiovascular disease: is abdominal obesity the missing link? Int. J. Obes. 27 (2003) S22–S24, https://doi.org/10.1038/sj.ijo.0802495.

[10] E. Forkert, T. Rendo-Urteaga, M.V. Nascimento-Ferreira, A. Ferreira de Moraes, L. Moreno, H. Barbosa de Carvalho, Abdominal obesity and cardiometabolic risk in children and adolescents, are we aware of their relevance? Nutrire 41 (2016) 15, https://doi.org/10.1186/s41110-016-0017-7.

[11] L. Wang, A.R. Folsom, J.H. Eckfeldt, Plasma fatty acid composition and incidence of coronary heart disease in middle aged adults: the Atherosclerosis Risk in Communities (ARIC) study, Nutr. Metab. Cardiovasc. Dis. 13 (2003) 256–266, https://doi.org/10.1016/s0939-4753(03)80029-7.

[12] K. Yamagishi, A.R. Folsom, L.M. Steffen, ARIC Study Investigators, Plasma fatty acid composition and incident ischemic stroke in middle-aged adults: the Atherosclerosis Risk in Communities (ARIC) study, Cerebrovasc. Dis. 36 (2013) 38–46, https://doi.org/10.1159/000351205.

[13] Z.D. Alsharari, U. Risérus, K. Leander, P. Sjögren, A.C. Carlsson, M. Vikström, et al., Serum fatty acids, desaturase activities and abdominal obesity - a population-based study of 60-year old men and women, PLoS One 12 (2017) e0170684, https://doi.org/10.1371/journal.pone.0170684.

[14] C. López-Gómez, C. Santiago-Fernández, S. García-Serrano, E. García-Escobar, C. Gutiérrez-Repiso, C. Rodríguez-Díaz, et al., Oleic acid protects against insulin resistance by regulating the genes related to the PI3K signaling pathway, J. Clin. Med. 9 (2020) 2615, https://doi.org/10.3390/jcm9082615.

[15] X. Palomer, J. Pizarro-Delgado, E. Barroso, M. Vázquez-Carrera, Palmitic and oleic acid: the yin and yang of fatty acids in type 2 diabetes mellitus, Trends Endocrinol. Metab. 29 (2018) 178–190, https://doi.org/10.1016/j.tem.2017.11.009.

[16] G. Rassias, M. Kestin, P.J. Nestel, Linoleic acid lowers LDL cholesterol without a proportionate displacement of saturated fatty acid, Eur. J. Clin. Nutr. 45 (1991) 315–320.

[17] R.P. Mensink, Effects of stearic acid on plasma lipid and lipoproteins in humans, Lipids 40 (2005) 1201–1205, https://doi.org/10.1007/s11745-005-1486-x.

[18] L.A. Gilmore, R.L. Walzem, S.F. Crouse, D.R. Smith, T.H. Adams, V. Vaidyanathan, et al., Consumption of high-oleic acid ground beef increases HDL-cholesterol concentration but both high- and low-oleic acid ground beef decrease HDL particle diameter in normocholesterolemic men, J. Nutr. 141 (2011) 1188–1194, https://doi.org/10.3945/jn.110.136085.

[19] T. Tholstrup, C. Ehnholm, M. Jauhiainen, M. Petersen, C.E. Høy, P. Lund, et al., Effects of medium-chain fatty acids and oleic acid on blood lipids, lipoproteins, glucose, insulin, and lipid transfer protein activities, Am. J. Clin. Nutr. 79 (2004) 564–569, https://doi.org/10.1093/ajcn/79.4.564.

[20] C.E. Kosmas, I. Martinez, A. Sourlas, K.V. Bouza, F.N. Campos, V. Torres, et al., High-density lipoprotein (HDL) functionality and its relevance to atherosclerotic cardiovascular disease, Drugs Context 7 (2018) 212525, https://doi.org/10.7573/dic.212525.

[21] M. Feng, M. Darabi, E. Tubeuf, A. Canicio, M. Lhomme, E. Frisdal, et al., Free cholesterol transfer to high-density lipoprotein (HDL) upon triglyceride lipolysis underlies the U-shape relationship between HDL-cholesterol and cardiovascular disease, Eur. J. Prev. Cardiol. 27 (2020) 1606–1616, https://doi.org/10.1177/2047487319894114.

[22] W.S. Davidson, R.A. Silva, S. Chantepie, W.R. Lagor, M.J. Chapman, A. Kontush, Proteomic analysis of defined HDL subpopulations reveals particle-specific protein clusters: relevance to antioxidative function, Arterioscler. Thromb Vasc Biol. 29 (2009) 870–876, https://doi.org/10.1161/ATVBAHA.109.186031.

[23] M. Ouimet, T.J. Barrett, E.A. Fisher, HDL and reverse cholesterol transport, Circ. Res. 124 (2019) 1505–1518, https://doi.org/10.1161/CIRCRESAHA.119.312617.

[24] M. Dobiášová, J.J. Frohlich, Advances in understanding of the role of lecithin cholesterol acyltransferase (LCAT) in cholesterol transport, Clin. Chim. Acta. 286 (1999) 257–271, https://doi.org/10.1016/s0009-8981(99)00106-0.

[25] P.J. Barter, H.B. Brewer Jr., M.J. Chapman, C.H. Hennekens, D.J. Rader, A.R. Tall, Cholesteryl ester transfer protein: a novel target for raising HDL and inhibiting atherosclerosis, Arterioscler. Thromb Vasc. Biol. 23 (2003) 160–167, https://doi.org/10.1161/01.atv.0000054658.91146.64.

[26] G. Assmann, H.U. Jabs, W. Nolte, H. Schriewer, Precipitation of LDL with sulphopolyanions: a comparison of two methods for LDL cholesterol determination, J. Clin. Chem. Clin. Biochem. 22 (1984) 781–785, https://doi.org/10.1515/cclm.1984.22.11.781.

[27] Expert Panel on Integrated Guidelines for Cardiovascular Health and Risk Reduction in Children and Adolescents; National Heart, Lung, and Blood Institute, Expert panel on integrated guidelines for cardiovascular health and risk reduction in children and adolescents: summary report, Pediatrics 128 (2011) S213–256, https://doi.org/10.1542/peds.2009-2107C.

[28] S.L. Aguilar-Espinosa, P. Mendoza-Espinosa, B. Delgado-Coello, J. Mas-Oliva, Lecithin cholesterol acyltransferase (LCAT) activity in the presence of Apo-AI-derived peptides exposed to disorder-order conformational transitions, Biochem. Biophys. Res.. Commun. 441 (2013) 469–475.

[29] G. Lepage, C.C. Roy, Direct transesterification of all classes of lipids in a one-step reaction, J. Lipid. Res. 27 (1986) 114–120.

[30] M. Rodríguez-Palmero, M.C. Lopez-Sabater, A.I. Castellote-Bargallo, M.C. De la Torre-Boronat, M. Rivero-Urgell, Comparison of two methods for the determination of fatty acid profiles in plasma and erythrocytes, J. Chromatogr. A. 793 (1998) 435–440.

[31] V. Hirschler, C. Aranda, M. de L. Calcagno, G. Maccalini, M. Jadzinsky, Can waist circumference identify children with the metabolic syndrome? Arch. Pediatr. Adolesc. Med. 159 (2005) 740–744, https://doi.org/10.1001/archpedi.159.8.740.

[32] A.O. Gobato, A.C. Vasques, M.P. Zambon, A. de A. Barros Filho, G. Hessel, Metabolic syndrome and insulin resistance in obese adolescents, Rev. Paul. Pediatr. 32 (2014) 55–62, https://doi.org/10.1590/s0103-05822014000100010.

[33] D.S. Freedman, M.K. Serdula, S.R. Srinivasan, G.S. Berenson, Relation of circumferences and skinfold thicknesses to lipid and insulin concentrations in children and adolescents: the Bogalusa heart study, Am. J. Clin. Nutr. 69 (1999) 308–317, https://doi.org/10.1093/ajcn/69.2.308.

[34] O. Ali, D. Cerjak, J.W. Kent, R. James, J. Blangero, Y. Zhang, Obesity, central adiposity and cardiometabolic risk factors in children and adolescents: a family-based study, Pediatr. Obes. 9 (2014) e58–e62, https://doi.org/10.1111/j.2047-6310.2014.218.x.

[35] A.S. Peçanha, A.M. Monteiro, F.M. Gazolla, I.R. Madeira, M.A.N. Bordallo, C.N.M. Carvalho, et al., Ultrasound as a method to evaluate the distribution of abdominal fat in obese prepubertal children and the relationship between abdominal fat and metabolic alterations, Radiol. Bras. 51 (2018) 293–296, https://doi.org/10.1590/0100-3984.2016.0230.

[36] B. Jin, H. Lin, J. Yuan, G. Dong, K. Huang, W. Wu, et al., Abdominal adiposity and total body fat as predictors of cardiometabolic health in children and adolescents with obesity, Front Endocrinol. (Lausanne) 11 (2020) 579, https://doi.org/10.3389/fendo.2020.00579.

[37] S.V. Galcheva, V.M. Iotova, Y.T. Yotov, S. Bernasconi, M.E. Street, Circulating proinflammatory peptides related to abdominal adiposity and cardiometabolic risk factors in healthy prepubertal children, Eur. J. Endocrinol. 164 (2011) 553–558, https://doi.org/10.1530/EJE-10-1124.

[38] G. Ponce, Sobrepeso, obesidad y factores de riesgo asociados en escolares de Comodoro Rivadavia, una alerta en el presente para evitar complicaciones futuras, Bioquímica y PatologíaClínica 76 (2012) 13–19.

[39] B. Mukherjee, C.M. Hossain, L. Mondal, P. Paul, M.K. Ghosh, Obesity and insulin resistance: an abridged molecular correlation, Lipid Insights 6 (2013) 1–11, https://doi.org/10.4137/LPI.S10805.

[40] J. Botton, B. Heude, P. Andre, J.L. Bresson, P. Ducimetiere, M.A. Charles, FLVS Study Group, Relationship between gamma-glutamyl transferase and fat mass in a general population of 8-17 years old children. The FLVS II study, Diabetes Metab 33 (2007) 354–359, https://doi.org/10.1016/j.diabet.2007.03.008.

[41] D.S. Bille, E. Chabanova, M. Gamborg, C.E. Fonvig, T.R. Nielsen, E. Thisted, et al., Liver fat content investigated by magnetic resonance spectroscopy in obese children and youths included in multidisciplinary

treatment, Clin. Obes. 2 (2012) 41–49, https://doi.org/10.1111/j.1758-8111.2012.00038.x.
[42] L. Meng, N. Luo, J. Mi, Impacts of types and degree of obesity on non-alcoholic fatty liver disease and related dyslipidemia in Chinese school-age children? Biomed. Environ. Sci. 24 (2011) 22–30, https://doi.org/10.3967/0895-3988.2011.01.003.
[43] V. Jain, M. Jana, B. Upadhyay, N. Ahmad, O. Jain, A.D. Upadhyay, et al., Prevalence, clinical & biochemical correlates of non-alcoholic fatty liver disease in overweight adolescents, Indian. J. Med. Res. 148 (2018) 291–301, https://doi.org/10.4103/ijmr.IJMR_1966_16.
[44] J. Revenga-Frauca, E.M. González-Gil, G. Bueno-Lozano, P. De Miguel-Etayo, P. Velasco-Martínez, J.P. Rey-López, et al., Abdominal fat and metabolic risk in obese children and adolescents, J. PhysiolBiochem. 65 (2009) 415–420, https://doi.org/10.1007/BF03185937.
[45] C.M. Velásquez-Rodríguez, M. Velásquez-Villa, L. Gómez-Ocampo, J. Bermúdez-Cardona, Abdominal obesity and low physical activity are associated with insulin resistance in overweight adolescents: a cross-sectional study, BMC Pediatr 14 (2014) 258, https://doi.org/10.1186/1471-2431-14-258.
[46] R.L. Wilensky, C.H. Macphee, Lipoprotein-associated phospholipase A (2) and atherosclerosis, Curr. Opin. Lipidol. 20 (2009) 415–420, https://doi.org/10.1097/MOL.0b013e3283307c16.
[47] M.H. Davidson, M.A. Corson, M.J. Alberts, J.L. Anderson, P.B. Gorelick, P.H. Jones, et al., Consensus panel recommendation for incorporating lipoprotein-associated phospholipase A2 testing into cardiovascular disease risk assessment guidelines, Am. J. Cardiol. 101 (2008) 51F–57F, https://doi.org/10.1016/j.amjcard.2008.04.019.
[48] A. Zalewski, C. Macphee, Role of lipoprotein-associated phospholipase A2 in atherosclerosis: biology, epidemiology, and possible therapeutic target, Arterioscler. Thromb Vasc. Biol. 2025 (2005) 923–931, https://doi.org/10.1161/01.ATV.0000160551.21962.a7.
[49] S.H. Law, M.L. Chan, G.K. Marathe, F. Parveen, C.H. Chen, L.Y. Ke, An updated review of lysophosphatidylcholine metabolism in human diseases, Int. J. Mol. Sci. 20 (2019) 1149, https://doi.org/10.3390/ijms20051149.
[50] L. Kheirandish-Gozal, M.F. Philby, Z. Qiao, A. Khalyfa, D. Gozal, endothelial dysfunction in children with obstructive sleep apnea is associated with elevated lipoprotein-associated phospholipase A2 plasma activity levels, J. Am. Heart Assoc. 6 (2017) e004923, https://doi.org/10.1161/JAHA.116.004923.
[51] M. Martin, L. Gaete, W. Tetzlaff, F. Ferraro, E. Lozano Chiappe, E.E. Botta, et al., Vascular inflammation and impaired reverse cholesterol transport and lipid metabolism in obese children and adolescents, Nutr. Metab. Cardiovasc. Dis. 32 (2022) 258–268, https://doi.org/10.1016/j.numecd.2021.09.025.
[52] S. Cook, R.E. Kavey, Dyslipidemia and pediatric obesity, Pediatr. Clin. North Am. 58 (2011) 1363–1673, https://doi.org/10.1016/j.pcl.2011.09.003, ix.
[53] G. Cohn, G. Valdes, D.M. Capuzzi, Pathophysiology and treatment of the dyslipidemia of insulin resistance, Curr. Cardiol. Rep. 3 (2001) 416–423, https://doi.org/10.1007/s11886-001-0059-0.
[54] C.E. Kosmas, G. Christodoulidis, J.W. Cheng, T.J. Vittorio, S. Lerakis, High-density lipoprotein functionality in coronary artery disease, Am. J. Med. Sci. 347 (2014) 504–508, https://doi.org/10.1097/MAJ.0000000000000231.
[55] G.E. Ronsein, J.W. Heinecke, Time to ditch HDL-C as a measure of HDL function? Curr. Opin. Lipidol. 28 (2017) 414–418, https://doi.org/10.1097/MOL.0000000000000446.
[56] S. Ebtehaj, E.G. Gruppen, S.J.L. Bakker, R.P.F. Dullaart, U.J.F. Tietge, HDL (High-Density Lipoprotein) cholesterol efflux capacity is associated with incident cardiovascular disease in the general population, Arterioscler. Thromb. Vasc. Biol. 39 (2019) 1874–1883, https://doi.org/10.1161/ATVBAHA.119.312645.
[57] H. Khalil, C. Murrin, M. O'Reilly, K. Viljoen, R. Segurado, J. O'Brien, et al., Total HDL cholesterol efflux capacity in healthy children - associations with adiposity and dietary intakes of mother and child, Nutr. Metab. Cardiovasc. Dis. 27 (2017) 70–77, https://doi.org/10.1016/j.numecd.2016.10.002.
[58] M.D. Barberio, L.J. Kasselman, M.P. Playford, S.B. Epstein, H.A. Renna, M. Goldberg, et al., Cholesterol efflux alterations in adolescent obesity: role of adipose-derived extracellular vesical microRNAs, J. Transl. Med. 17 (2019) 232, https://doi.org/10.1186/s12967-019-1980-6.
[59] J. Härdfeldt, M. Cariello, S. Simonelli, A. Ossoli, N. Scialpi, M. Piglionica, et al., Abdominal obesity negatively influences key metrics of reverse cholesterol transport, Biochim. Biophys. Acta Mol. Cell. Biol. Lipids. 1867 (2022) 159087, https://doi.org/10.1016/j.bbalip.2021.159087.
[60[S. Bonafini, A. Giontella, A. Tagetti, I. Bresadola, R. Gaudino, P. Cavarzere, et al., Fatty acid profile and desaturase activities in 7-10-year-old children attending primary school in verona south district: association between palmitoleic acid, SCD-16, indices of adiposity, and blood pressure, Int. J. Mol. Sci. 21 (2020) 3899, https://doi.org/10.3390/ijms21113899.
[61] T. Okada, N. Furuhashi, Y. Kuromori, M. Miyashita, F. Iwata, K. Harada, Plasma palmitoleic acid content and obesity in children, Am. J. Clin. Nutr. 82 (2005) 747–750, https://doi.org/10.1093/ajcn/82.4.747.
[62] T. Marosvölgyi, É. Szabó, É. Erhardt, D. Molnár, T. Decsi, High contribution of palmitoleic acid to plasma lipid classes in obese children with and without metabolic risk factors related to obesity, J. Ped. Gastr. Nutr. 42 (2006) E83.
[63] M.C. Hua, H.M. Su, M.W. Lai, T.C. Yao, M.H. Tsai, S.L. Liao, et al., Palmitoleic and dihomo-γ-linolenic acids are positively associated with abdominal obesity and increased metabolic risk in children, Front. Pediatr. 9 (2021) 628496, https://doi.org/10.3389/fped.2021.628496.
[64] J.C. Aristizabal, L.I. González-Zapata, A. Estrada-Restrepo, J. Monsalve-Alvarez, S.L. Restrepo-Mesa, D. Gaitán, Concentrations of plasma free palmitoleic and dihomo-gamma linoleic fatty acids are higher in children with abdominal obesity, Nutrients 10 (2018) 31, https://doi.org/10.3390/nu10010031.
[65] M.T. Flowers, The delta 9 fatty acid desaturation index as a predictor of metabolic disease, Clin. Chem. 55 (2009) 2071–2073, https://doi.org/10.1373/clinchem.2009.135152.
[66] Y.E. Zhou, G.M. Egeland, S.J. Meltzer, S. Kubow, The association of desaturase 9 and plasma fatty acid composition with insulin resistance-associated factors in female adolescents, Metabolism 58 (2009) 158–166, https://doi.org/10.1016/j.metabol.2008.09.008.
[67] J. Korbecki, K. Bajdak-Rusinek, The effect of palmitic acid on inflammatory response in macrophages: an overview of molecular mechanisms, Inflamm. Res. 68 (2019) 915–932, https://doi.org/10.1007/s00011-019-01273-5.
[68] A. Piano, D. Masquio, A. Dâmaso, L. Oyama, D. Estadella, A. Chamas, et al., Serum myristic fatty acid negatively correlates with anti-inflammatory adiponectin/leptin ratio in obese adolescents: effects of long- term therapy, O Mundo da Saúde 40 (2017) 537–554, https://doi.org/10.15343/0104-7809.201740A537554.
[69] V. Saraswathi, N. Kumar, W. Ai, T. Gopal, S. Bhatt, E.N. Harris, et al., Myristic acid supplementation aggravates high fat diet-induced adipose inflammation and systemic insulin resistance in mice, Biomolecules 12 (2022) 739, https://doi.org/10.3390/biom12060739.
[70] J.K. Innes, P.C. Calder, Omega-6 fatty acids and inflammation, Prostaglandins Leukot Essent Fatty Acids 132 (2018) 41–48, https://doi.org/10.1016/j.plefa.2018.03.004.
[71] P. de Pablo, D. Romaguera, H.L. Fisk, P.C. Calder, A.M. Quirke, A.J. Cartwright, et al., High erythrocyte levels of the n-6 polyunsaturated fatty acid linoleic acid are associated with lower risk of subsequent rheumatoid arthritis in a southern European nested case-control study, Ann. Rheum. Dis. 77 (2018) 7981–7987, https://doi.org/10.1136/annrheumdis-2017-212274.
[72] D. Tricò, A. Di Sessa, S. Caprio, N. Chalasani, W. Liu, T. Liang, et al., Oxidized derivatives of linoleic acid in pediatric metabolic syndrome: is their pathogenic role modulated by the genetic background and the gut microbiota? Antioxid. Redox. Signal. 30 (2019) 241–250, https://doi.org/10.1089/ars.2017.7049.
[73] E. Demetz, A. Schroll, K. Auer, C. Heim, J.R. Patsch, P. Eller, et al., The arachidonic acid metabolome serves as a conserved regulator of cholesterol metabolism, Cell. Metab. 20 (2014) 787–798, https://doi.org/10.1016/j.cmet.2014.09.004.
[74] Y. Wang, J.F. Oram, Unsaturated fatty acids inhibit cholesterol efflux from macrophages by increasing degradation of ATP-binding cassette transporter A1, J. Biol. Chem. 277 (2002) 5692–5697, https://doi.org/10.1074/jbc.M109977200.
[75] K.F. Austen, The role of arachidonic acid metabolites in local and systemic inflammatory processes, Drugs 33 (1987) 10–17, https://doi.org/10.2165/00003495-198700331-00004.
[76] N.L. Spartano, S. Lamon-Fava, N.R. Matthan, M.S. Obin, A.S. Greenberg, A.H. Lichtenstein, Linoleic acid suppresses cholesterol efflux and ATP-binding cassette transporters in murine bone marrow-derived macrophages, Lipids 49 (2014) 415–422, https://doi.org/10.1007/s11745-014-3890-y.
[77] I. Kämmerer, R. Ringseis, R. Biemann, G. Wen, K. Eder, 13-hydroxy linoleic acid increases expression of the cholesterol transporters ABCA1, ABCG1 and SR-BI and stimulates apoA-I-dependent cholesterol efflux in RAW264.7 macrophages, Lipids Health Dis 30 (2011) 222, https://doi.org/10.1186/1476-511X-10-222.

[78] L. Hsu, Z. Wen, H. Chen, H. Lin, C. Chiu, H. Wu, Evaluation of the anti-inflammatory activities of 5,8,11-cis-eicosatrienoic acid, Food Nutr. Sci. 4 (2013) 113–119, https://doi.org/10.4236/fns.2013.49A1018.

[79] A.M. Bérard, H. Dabadie, A. Palos-Pinto, M.F. Dumon, M. Darmon, Reduction of dietary saturated fatty acids correlates with increased plasma lecithin cholesterol acyltransferase activity in humans, Eur. J. Clin. Nutr. 58 (2004) 881–887, https://doi.org/10.1038/sj.ejcn.1601890.

[80] C. Vaysse-Boué, H. Dabadie, E. Peuchant, P. Le Ruyet, F. Mendy, H. Gin, et al., Moderate dietary intake of myristic and alpha-linolenic acids increases lecithin-cholesterol acyltransferase activity in humans, Lipids 42 (2007) 717–722, https://doi.org/10.1007/s11745-007-3074-0.

[81] G. Zong, Y. Li, A.J. Wanders, M. Alssema, P.L. Zock, W.C. Willett, et al., Intake of individual saturated fatty acids and risk of coronary heart disease in US men and women: two prospective longitudinal cohort studies, BMJ 355 (2016) i5796, https://doi.org/10.1136/bmj.i5796.

[82] B. Papotti, J.C. Escolà-Gil, J. Julve, F. Potì, I. Zanotti, Impact of dietary lipids on the reverse cholesterol transport: what we learned from animal studies, Nutrients 39 (2021) 2643, https://doi.org/10.3390/nu13082643.

[83] L.B. Bockus, M.L. Biggs, H.T.M. Lai, M.C. de Olivera Otto, A.M. Fretts, B. McKnight, et al., Assessment of plasma phospholipid very-long-chain saturated fatty acid levels and healthy aging, JAMA Netw. Open. 4 (2021) e2120616, https://doi.org/10.1001/jamanetworkopen.2021.20616.

[84] H.K. Chung, Y. Cho, H.J. Do, K. Oh, W.K. Seo, M.J. Shin, Plasma phospholipid arachidonic acid and lignoceric acid are associated with the risk of cardioembolic stroke, Nutr. Res. 35 (2015) 1001–1008, https://doi.org/10.1016/j.nutres.2015.09.007.

[85] M. Liu, L.S. Zuo, T.Y. Sun, Y.Y. Wu, Y.P. Liu, F.F. Zeng, et al., Circulating very-long-chain saturated fatty acids were inversely associated with cardiovascular health: a prospective cohort study and meta-analysis, Nutrients 12 (2020) 2709, https://doi.org/10.3390/nu12092709.

[86] W. Qureshi, I.D. Santaren, A.J. Hanley, S.M. Watkins, C. Lorenzo, L.E. Wagenknecht, Risk of diabetes associated with fatty acids in the de novo lipogenesis pathway is independent of insulin sensitivity and response: the Insulin Resistance Atherosclerosis Study (IRAS), BMJ Open Diabetes Res. Care 7 (2019) e000691, https://doi.org/10.1136/bmjdrc-2019-000691.

[87] Y. Takenouchi, Y. Seki, S. Shiba, K. Ohtake, K. Nobe, K. Kasono, Effects of dietary palmitoleic acid on vascular function in aorta of diabetic mice, BMC Endocr. Disord. 22 (2022) 103, https://doi.org/10.1186/s12902-022-01018-2.

[88] C.O. Souza, A.A. Teixeira, L.A. Biondo, L.S. Silveira, P.C. Calder, J.C. Rosa Neto, Palmitoleic acid reduces the inflammation in LPS-stimulated macrophages by inhibition of NFκB, independently of PPARs, Clin. Exp. Pharmacol. Physiol. 44 (2017) 566–575, https://doi.org/10.1111/1440-1681.12736.

[89] D. Tricò, A. Mengozzi, L. Nesti, M. Hatunic, R. Gabriel Sanchez, T. Konrad, et al., Circulating palmitoleic acid is an independent determinant of insulin sensitivity, beta cell function and glucose tolerance in non-diabetic individuals: a longitudinal analysis, Diabetologia 63 (2020) 206–218, https://doi.org/10.1007/s00125-019-05013-6.