Lipid Tetrad Index (LTI) and Lipid Pentad Index (LPI) in Healthy Subjects

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Abstract

Background: The prevalence of cardiovascular disease (CVD) has increased steadily in recent years. Literature data show that about 35% of atherosclerotic events occur in the absence of classic risk factors, requiring a broader assessment of the individual to better characterize the risk. Lipid Tetrad Index (LTI) and Lipid Pentad Index (LPI) constitute a new and efficient evaluation of the lipid profile and CVD risk.

Objective: This study assessed LTI and LPI in undergraduate students, seeking to establish the parameters of these indices in healthy subjects and correlate them with the conventional lipid profile.

Methods: The study included 110 students, 48 (44%) males and 62 (56%) females, mean age 20.9 ± 1.7. Apolipoprotein-AI, apolipoprotein B, total cholesterol, lipoprotein(a), triglycerides, high-density lipoprotein (HDL) and low-density lipoprotein (LDL) were assessed, using specific diagnostic methods. LTI and LPI indices were calculated using the equations LTI = [total cholesterol x triglycerides x lipoprotein(a) / HDL] and LPI = [total cholesterol x triglycerides x lipoprotein(a) x apolipoprotein B/apolipoprotein-AI], respectively.

Results: LTI and LPI values were significantly higher in females compared to males. As for the other parameters, there were significant differences between males and females only regarding total cholesterol, HDL and apolipoprotein-AI. There were significant and positive correlations between LDL and LTI and between LDL and LPI.

Conclusions: Findings indicate that both LTI and LPI were associated with LDL, a parameter not used to calculate lipid indices and widely used in clinical practice for cardiovascular risk assessment (Arq Bras Cardiol. 2013;100(4):322-327).

Keywords: Risk Factors; Lipids; Cardiovascular Diseases; Students.

Introduction

The high prevalence of cardiovascular disease (CVD) in Brazil is due to genetic factors, as well as environmental events that can lead to the onset and progression of atheroma plaques[1,2]. Unbalanced diet, smoking, physical inactivity, diabetes and high cholesterol are factors that tend to greatly increase the risk of developing CVD[2].

Literature data have consolidated the association between plasma levels of total cholesterol, LDL and triglycerides and the occurrence of these events[3-5], with hypercholesterolemia being an important factor in approximately 50% of cases[6]. However, many cardiovascular events occur in normolipidemic subjects[7,8]. De Backer et al[9] reported that one third of cases of acute myocardial infarction (AMI) were observed in patients with intermediate risk status. New biomarkers have been evaluated in an attempt to elucidate factors that lead to these events and among these, lipoprotein (a) [Lp (a)], a genetic variant of LDL that has apolipoprotein (a) linked by disulfide bonds to apolipoprotein B (apoB)[10,11].

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In this context, the need to search for new methods of analysis has led to studies that more comprehensively describe patients' actual propensity to develop CVD, overcoming the analysis commonly based on individual lipid particles. The lipid tetrad (LTI) and pentad (LPI) indices have recently been described as a new form of assessment of lipid profiles, which have been analyzed in some populations[12,13]. A characteristic of these new indices is distinguished by the broad approach of atherogenic and non-atherogenic lipid particles, resulting in a single value. Based on the conventional lipid profile and the emerging risk factors such as Lp (a), apolipoprotein AI (apoA-I) and apoB, the LTI and LPI appear as models in global risk assessment, considering the multifactorial nature of CVD.

The purpose of this study was to establish parameters of these new indices in healthy individuals from the Brazilian population, given that, to date, no study has been carried out in this population, which consists of different ethnic groups[14]. We also proposed to evaluate the correlation between the values of these new indices with those traditionally used, in addition to assessing other risk factors in this population.

Methods

After project approval by the Ethics Committee on Human Research of the University, the study was carried out between the months of May and November 2011, in supposedly healthy individuals, all undergraduate students aged between 18 and 25 years. The volunteers signed an informed consent form agreeing to participate in the study and the purposes and objectives of the research were specified. A questionnaire was applied, which included data such as age, year of graduation, attended period, family history of CVD, smoking and lifestyle. Additionally, weight, height, waist measurement and blood pressure were measured. Assignments followed the guidelines of the Brazilian Society of Cardiology (SBC)[2].

After a 12-hour fast, blood samples were obtained from the volunteers using Vacutainer® tubes (Becton & Dickson) without anticoagulant. These samples were centrifuged at 2500 rpm for 15 minutes and the serum was divided in aliquots, labeled and stored at -70 °C until the tests were performed.

The enzymatic colorimetric methods Cholesterol Monoreagent K082 and Triglycerides Monoreagent K117 by Bioclin® were used for the measurement of total cholesterol and triglycerides, respectively. HDL and LDL were determined by enzymatic elimination method HDL Cholesterol Direct K071 and LDL Cholesterol Direct K088, respectively, by Bioclin®. Measurements of Apo A-I and Apo B were performed using the immunoturbidimetry methods of Apolipoprotein AI Turbidimetry and Apolipoprotein B Turbidimetry by Biotecnica®, respectively. Lp (a) was also obtained by the immunoturbidimetry method, using the *In Vitro* Turbidimetric Lipoprotein (a) diagnostic set.

The tests were performed in a Cobas Mira Plus device, using commercial control sera to verify assay performance. Control liquid (Spin React) was used for the measurement of Lp (a). LTI was obtained by the equation LTI = Total Cholesterol x Triglycerides x Lp (a)/HDL and LPI = Total Cholesterol x Triglycerides x Lp (a) x apoB/apoA-I [12,13].

The minimum sample size was defined by the coefficient of variation obtained for Lp (a) measurement (103%), considering ten percent variation around the mean, reaching a minimum of sixteen individuals in each group to demonstrate the possible statistical differences with a significance level of 5%. Student's *t* tests for normally distributed variables and Mann-Whitney test for nonparametric variables were used for the statistical analysis. For the analysis of the correlation, Pearson's correlation test was used. The level of significance was set at 5%. The software programs Sigma Stat® release 1.0 and Prism® release 3.0 were used to perform the analysis and plot graphs, respectively.

Results

Table 1 shows the study population and its general characteristics according to gender. The study included 110 students, 48 males (44%) and 62 females (56%).

According to the guidelines of the Brazilian Society of Cardiology[2] for total cholesterol (TC), LDL, triglycerides (TG) and HDL there were no mean values outside the range recommended for this population in general. The same was observed for the values regarding each gender. However, significant differences wee observed between genders for TC, HDL, Lp (a) and apoA-I considering the level of significance used, which were higher in females. No significant differences were observed regarding other parameters such as LDL and apoB, which had p values close to the threshold. Of the female students, a significant number had some type of dyslipidemia, with this prevalence being statistically significant in relation to male students. Among the risk factors analyzed, the most prevalent were sedentary lifestyle, present in 68.1% of the students, followed by family history, at 59.1%. Other data did not differ significantly and/or did not show any relevance.

The tetrad and pentad lipid indices are sown in Table 1 and in Charts 1 and 2 respectively, also being shown as logarithmic values. LTI was higher in the female population, which is statistically demonstrated ($p = 0.024$). Chart 1 illustrates this visually observed difference through the horizontal line that represents the mean of these values for both genders. The same was observed for LPI ($p = 0.007$), also demonstrating higher levels in women and shown in Chart 2.

Another analysis was performed to evaluate possible associations between LPI and LTI and other parameters that are not included in their calculation. Thus, we found positive and significant correlations between LTI and LDL ($r = 0.49$, $p < 0.001$) and between LPI and LDL ($r = 0.56$, $p < 0.001$).

Discussion

The lack of alterations in the traditional lipid levels can be attributed to the studied population, which consisted exclusively of undergraduate, young and presumably healthy students. Some volunteers had dyslipidemia, 28.2% of the total, which can be explained by individual genetic predisposition. However, normolipidemic individuals predominated in the studied population, both in males and females, confirming a similar study carried out by Coelho et al. (2005) in university students[15].

When performing the analysis taking gender into account, total cholesterol, HDL, Lp (a) and apoA-I levels were higher in women, when compared to men. There is disagreement in the literature regarding the differences or not in lipid profile of men and women, given the fluctuations in lipid levels at different stages of the female menstrual cycle due to the constant metabolic changes caused by estrogen[16-18]. It is noteworthy the fact that both HDL and apoA-I, considered antiatherogenic and protective factors against CVD, and TC and Lp (a), considered atherogenic factors that may predispose to cardiovascular events, were high in women, when compared to men (Table 1). These findings allow us to conclude that the evaluation of individual parameters can often become complicated in clinical practice and an overall assessment proposed by LTI and LPI can be a tool to aid patient assessment.

Even though SBC[2] does not recommend the routine measurement of apolipoproteins A-I and B and Lp (a), the conventional lipid profile (TC and fractions) may not demonstrate coronary risk, as there were no significant differences in LDL levels between men and women, and women had significantly higher mean levels of HDL than men. On the other hand, the results obtained for LTI and LPI were also significantly higher in women (Table 1). These findings reinforce the idea that the

Table 1 – Characteristics of the study group and results

Parameter	Total	Male sex	Female sex	p
n	110 (100%)	48 (44%)	62 (56%)	---
Age (years)	20.9 ± 1.7	21.0 ± 1.8	20.8 ± 1.6	0.607
BMI (Kg/m²)	22.3 ± 2.7	22.9 ± 3.1	21.9 ± 2.3	0.055
Waist circumference (cm)	76.9 ± 7.6	81.2 ± 7.3	73.8 ± 6.2	< 0.001(*)
SBP (mmHg)	111 ± 9.4	113 ± 8 110 (110 – 120)	109 ± 10 110 (100 – 110)	< 0.001(†)
DBP (mmHg)	70 ± 9	72 ± 9	69 ± 9	0.099
Diabetes mellitus	0	0	0	---
Smoking	6 (5.5%)	4 (8.3%)	2 (3.1%)	0.227(‡)
Arterial hypertension	1 (0.9%)	1 (2.1%)	0	---
Sedentary life style	75 (68.1%)	33 (68.8%)	42 (67.7%)	0.538(§)
Family history	65 (59.1%)	30 (62.5%)	35 (56.4%)	0.329(§)
Dyslipidemias	31 (28.2%)	8 (16.7%)	23 (37.1%)	0.015(*)
Total cholesterol (mg/dL)	168 ± 34	156 ± 33	178 ± 32	< 0.001(*)
Triglycerides (mg/dL)	81 ± 46 68 (50 – 98)	74 ± 39 66 (47 – 80)	86 ± 50 70 (53 – 105)	0.149
HDL (mg/dL)	55 ± 15 53 (44 – 62)	49 ± 12 47 (41 – 54)	59 ± 14 58 (50 – 65)	< 0.001(†)
LDL (mg/dL)	97 ± 29	91 ± 29	101 ± 28	0.092
Lipoprotein (a) (mg/dL)	27.3 ± 31.5 16.4 (5.5 – 34.7)	19.2 ± 21.0 11.3 (3.1 – 30.7)	33.7 ± 36.6 20.9 (6.5 – 42.3)	0.017(†)
Apolipoprotein A-I (mg/dL)	139 ± 32 137 (121 – 159)	128 ± 22 124 (115 – 137)	148 ± 36 151 (129 – 173)	< 0.001(*)
Apolipoprotein B (mg/dL)	80 ± 20	76 ± 19	83 ± 20	0.073
Lipid tetrad index (LTI)	7,989 ± 12.064 3,628 (1.016 – 8.360)	5,850 ± 9.232 2,721 (539 – 5.107)	9,645 ± 13.712 4,868 (1.779 – 11.544)	0.018(†)
Log LTI	3,50 ± 0,63	3,35 ± 0,63	3,62 ± 0,61	0,024(*)
Lipid pentad index (LPI)	312,607 ± 598.936 107,541 (32.029 – 251.033)	193,125 ± 174.160 76,706 (15.352 – 159.882)	405,109 ± 716,382 137,050 (53,889 – 382,109)	0,007(†)
Log LPI	4.99 ± 0.69	4.79 ± 0.66	5.15 ± 0.66	0,007(*)

Data are presented as mean ± standard deviation for continuous variables and number of participants and percentage for categorical variables. The variables without normal distribution were also presented as median and interquartile difference. n: sample size, BMI: body mass index, SBP: systolic blood pressure, DBP: diastolic blood pressure, p: probability for hypothesis testing. (*) Student's T-test (variable with normal distribution), (†) Mann-Whitney test (nonparametric variable), (‡) Fisher's Exact Test, (§) Chi-square.

conventional lipid profile, in many cases, may not offer an actual estimation of coronary risk. Thus, when this profile is normal, it does not necessarily rule out the risk due to dyslipidemia. Consequently, the introduction of laboratory tests related to the unconventional lipid profile would be extremely desirable in search of the real coronary risk estimate, as an addition to all other traditionally investigated risk factors.

The higher number of dyslipidemia cases among women alert to the possible development of atherosclerotic plaques and all its consequences in this group of individuals in the future. Some studies point to a sedentary lifestyle among students as a possible aggravating factor for the increase in lipid levels, especially noticeable in women[15,19]. A non-modifiable risk factor observed in 59.1% (Table 1) of the students was the family history for CVD, also demonstrated in other studies[15,19,20]. Considering all these data, they allow us to infer that this large proportion of students with family history of CVD did not imply in increased health awareness, as a modifiable factor such as physical inactivity was strongly present in this population (68.1%).

LTI was described by Enas et al[12] as a new way to assess cardiovascular risk. Characterized by the multiplication of three atherogenic particles, which are total cholesterol, triglycerides and Lp (a), and the division of this product by the non-atherogenic HDL particle, the index describes the overall lipid profile of patients. This index has been compared between healthy control subjects and those with coronary artery disease, with healthy subjects showing much lower levels[12,21].

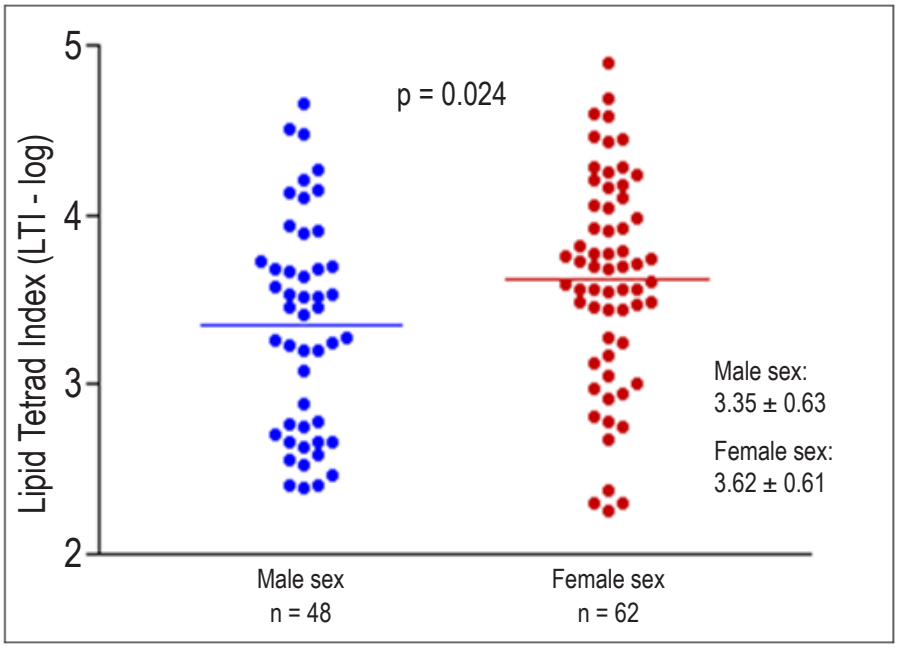

Chart 1 – *Distribution of LTI values according to gender.*
LTI values calculated for men and women were compared, focusing on the mean of each group. Note that the mean in females was significantly higher compared to males. n: sample size, p: probability for hypothesis testing.

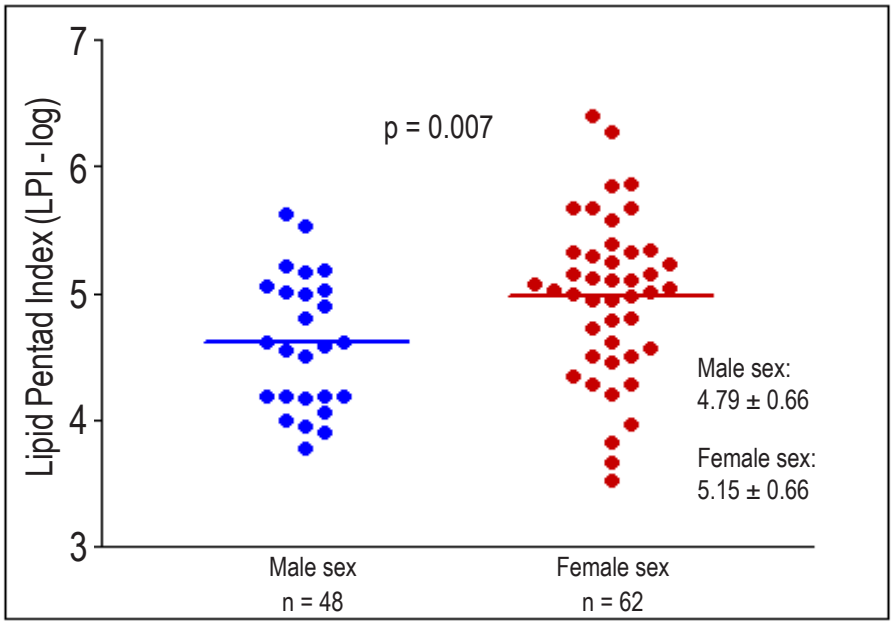

Chart 2 – *Distribution of LPI values according to gender.*
LPI values calculated for both genders were compared, focusing on the mean of each group. Note that the mean in females was significantly higher than that in males. n: sample size, p: probability for hypothesis testing.

LPI comprises the product of four atherogenic particles, namely total cholesterol, triglycerides, Lp (a) and ApoB, divided by Apo A-1, which is non-atherogenic. Initially described by Das et al[13] as a better way to discriminate cases of CAD in relation to LTI in children and adolescents, LPI encompasses both alterations in the traditional lipid profile and emerging factors, such as Lp (a), Apo A-1 and Apo B.

Both LTI as LPI levels were higher in women. The fact that there have been few records in the literature on these new indices makes the present work a pilot in the description of possible gender differences. Additional investigations are necessary to clarify this principle, considering the metabolic changes observed due to the female menstrual cycle[18]. However, even in these observations, the occurrence of higher levels in the female students does not rule out their increased propensity to a future risk of developing CVD and actions that may minimize this predisposition should be undertaken.

In an attempt to assess the association between levels commonly described in the literature with LTI and LPI, correlation tests were performed. As the indices comprise some of these traditional particles, correlations between them and their component particles do exist, but would not be credited. The correlation observed between LTI and LDL (49%) and between LPI and LDL (56%) establishes a connection that may be the way to validate these new indices. It is noteworthy the fact that LDL is not part of either of the two equations used for calculating LTI and LPI. Although the degree of the correlations has not been high from the mathematical point of view, these cannot be overlooked clinically, given the great differences in lipid levels among individuals, and even regarding gender, as mentioned before. Moreover, the multiparametric characteristic in the constitution of LTI and LPI supports this degree of correlation.

The LTI and LPI values found in the study population may contribute to establish reference values of these indices in the Brazilian population, as there are few studies in the literature and none, to date, for this purpose. The young population is a model for the acquisition of these data as it supposedly includes healthy individuals. However, the values of these indices cannot be used as yet as parameters indicative of dyslipidemia as, so far, there are no references described in the literature, one of the reasons why this study becomes appropriate. Additional studies involving a larger number of individuals are needed for a better description of these parameters, as well as the occurrence of a possible difference between genders.

Author contributions

Conception and design of the research, Statistical analysis, Obtaining funding and Critical revision of the manuscript for intellectual content: Lima LM; Acquisition of data and Writing of the manuscript: Morais CAS, Oliveira SHV; Analysis and interpretation of the data: Morais CAS, Oliveira SHV, Lima LM.

Potential Conflict of Interest

No potential conflict of interest relevant to this article was reported.

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Study Association

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