Assessment of serum Lipoprotein(a) status during type 2 diabetes mellitus

Abstract

Introduction: Lipoprotein (a) is made up of an atherogenic LDL lipoparticle and a potentially thrombogenic apoprotein a and is therefore responsible for cardiovascular disease. The objective of this study is to evaluate serum lipoprotein (a) status and to investigate the correlation of elevated serum lipoprotein (a) levels with other cardiovascular risk factors in type 2 diabetics.

Material and methods: This is a case-control study involving 82 patients, 37 type 2 diabetic patients and 45 non-diabetic control subjects. Sociodemographic data were collected and each patient underwent routine lipid assessment and lipoprotein (a) testing.

Results : The prevalence of hyperlipoproteinemia (a) is 17.8% in control subjects and 29.7% in type 2 diabetics. HDL cholesterolemia is significantly higher in controls than in type 2 diabetics (p =0.028) while LDL cholesterol and serum lipoprotein (a) levels are higher in type 2 diabetics than in controls with a statistically significant difference (p=0.025 and p=0.026 respectively). The mean lipoprotein (a) values of 0.36±0.34 g/l in women are higher than those of male subjects which are 0.28±0.20 g/l (p=0.171). Mean serum lipoprotein (a) levels of 0.39±0.32 g/l in type 2 diabetics are significantly higher than those of controls which are 0.25±0.21 g/l (p= 0.026). Plasma concentrations of lipoprotein (a) vary with age and appear to be increased beyond the age of 45. There is no correlation between lipoprotein (a) and other cardiovascular risk factors.

Conclusion : Hyperlipoproteinemia (a) is common in type 2 diabetics and women have the highest plasma levels. Serum lipoprotein (a) concentrations are not correlated with other cardiovascular risk factors and therefore constitute an independent risk factor.

Key words: lipoprotein (a), cardiovascular risk factors, routine lipid profile, type 2 diabetes.

Introduction

Patients with type 2 diabetes mellitus (T2DM) are at high risk of cardiovascular disease, which remains the leading cause of morbidity and mortality worldwide and dyslipidemia is a major cardiovascular risk factor in these patients [**1**]. Lipoprotein(a) or Lp(a) is a glycoprotein synthesized in the liver, formed from a molecule analogous to low-density lipoprotein (LDL), associated with an apolipoprotein a (apo a) molecule. Like LDL, Lp(a) is made up of a protein part, apolipoprotein B100 (apo B100) linked to apo a by a disulfide bridge and a lipid part rich in cholesterol, cholesterol esters and in phospholipids [**2, 3**]. Lp(a) consists of an atherogenic LDL lipoparticle and a potentially thrombogenic apo a and its causal role as a risk factor for cardiovascular disease has been well established [**3, 4**]. A subtype of LDL, it therefore constitutes a lipoprotein of interest and an independent cardiovascular risk factor little evaluated in Africa and in Senegal in particular. Thus, this study aims to determine the serum Lp(a) status and to investigate the correlation of elevated serum Lp(a) levels with other cardiovascular risk factors in type 2 diabetic subjects.

Material and methods

Population and study setting

This is a case-control study carried out in the commune of Ziguinchor (Senegal) over a period of 4 months, involving 82 patients, 37 type 2 diabetic patients followed at the antidiabetic service of the Regional Hospital Center of Ziguinchor and 45 non-diabetic control subjects, free from moderate fasting hyperglycemia and without glucose intolerance, recruited from a private medical facility.

Inclusion and exclusion criteria

Included in the study were type 2 diabetics and controls who agreed to participate after free and informed consent. The exclusion criteria were subjects presenting a pathology that could interfere with the serum concentration of lipoprotein (a) such as renal pathology, hepatic pathology, dysthyroidism, acute inflammation or chronic inflammatory pathology.

Sociodemographic and clinical data

Age and sex were determined. Other cardiovascular risk factors such as smoking, alcohol consumption, high blood pressure (hypertension) and obesity were collected. An age strictly greater than 50 years in men and 60 years in women was also defined as a cardiovascular risk factor.

Biological investigations

A venous blood sample was taken from the subjects thus selected (patients and controls) fasting for 8 to 12 hours, at the elbow crease after application of a tourniquet and the blood was collected in a dry tube without anticoagulant for the needs for routine lipid assessment and Lp(a) testing. The blood samples obtained were then centrifuged and the serums were collected and then frozen immediately at -20°C until the time of assay.For the purposes of the assay, the samples were thawed at room temperature then homogenized using a blender. The determination of total cholesterol, HDL cholesterol and triglycerides was carried out by enzymatic method (Biosystem®, Barcelona, Spain) and that of Lp(a) by immunoturbidimetric method (Abbott Diagnostics®, Sligo, Ireland). The concentration of LDL cholesterol was using the Friedewald formula while respecting the condition of its applicability, namely a triglyceridemia lower than 3.5 g/l.

Data analysis

The data were entered into Excel 2013 and analyzed using R software in version 4.3.1. The Pearson correlation test was determined to study the association

between Lp(a) and other cardiovascular risk factors. The Student t test was used to compare the different average concentrations obtained. We considered a significant difference for a $p<0.05$.

Results

The study population, made up of 37 T2DM and 45 controls, is characterized by a male predominance. High serum levels of Lp(a) were found in 29.7% of T2DM and with a prevalence of dyslipidemia of 51.3%. The other clinical and sociodemographic characteristics of the study population are summarized in Table 1.

Table 1 : Socio-demographic and clinical characteristics of study population

Parameters	**T2D n=37 (%)**	**Controle n=45 (%)**
Age (years)	50.9±7.7	41±7.9
Men	20 (54.1%)	27 (60%)
Smoking	0 (0%)	2 (4,4%)
Alcohol consumption	0 (0%)	0 (0%)
Obesity	5 (13,5%)	12 (26,7%)
HBP (mmHg)	8 (21,6%)	6 (13,3%)
Dyslipidemia	19 (51.3%)	8 (17.8%)
High levels of Lp(a)	11 (29.7%)	8 (17.8%)

The lipid profile parameters (table 2) show that HDL cholesterol levels are significantly higher in controls than T2D patients ($p=0.028$) while LDL cholesterol levels and serum Lp(a) levels are higher in T2D patients than in controls with a statistically significant difference ($p=0.025$ and $p=0.026$ respectively).

Table 2 : Biological characteristics of study population

Parameters	**T2D (n=37)**	**Controle (n=45)**	**p-value**
Total cholesterolemia (g/l)	1.81±0.54	1.81±0.42	0.993
HDL cholesterolemia (g/l)	0.44±0.31	0.56±0.16	**0.028**
LDL cholesterolemia (g/l)	1.18±0.49	1.05±0.44	0.203
LDL/HDL	4.83±6.97	2.32±2.16	**0.025**
Triglyceridemia (g/l)	0.95±0.44	0.98±0.51	0.762
Triglyceridemia/HDL	5.47±10.5	2.03±1.59	0.141
Lp(a) (g/l)	0.39±0.32	0.25±0.21	**0.026**

Figure 1 shows a variation in Lp(a) concentrations in the study population depending on sex. The average values of Lp(a), 0.36±0.34 g/l in women are higher than those of male subjects which are 0.28±0.20 g/l with a non-significant difference (p=0.171). Mean serum Lp(a) levels are 0.39±0.32 g/l in T2DM and 0.25±0.21 g/l in controls. Their difference is statistically significant (p=0.026).

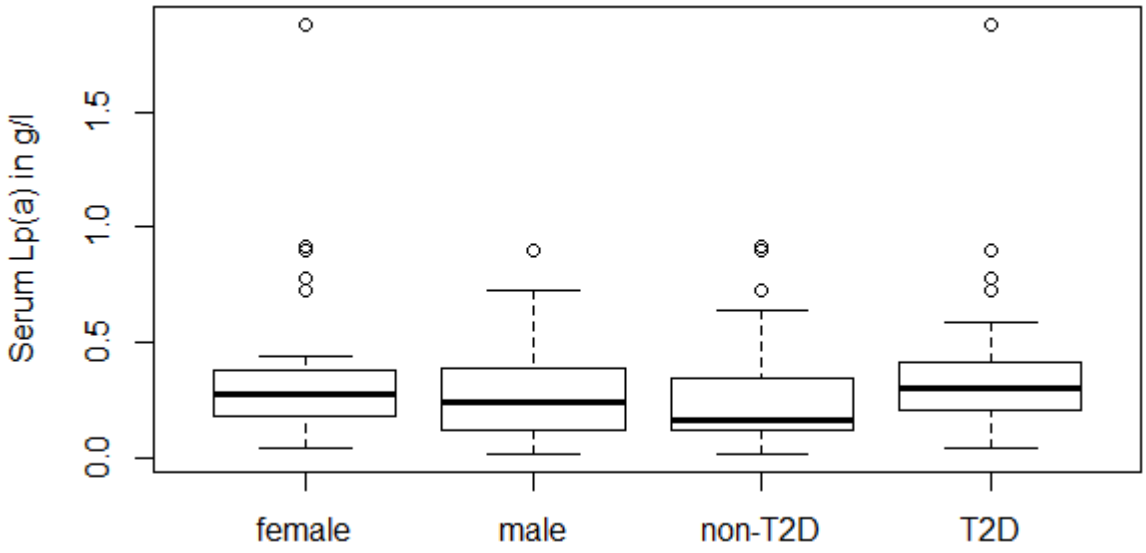

Figure 1 : Variation in serum Lp(a) levels of the study population according to sex

Serum Lp(a) concentrations vary with age and appear to be increased beyond the age of 45 (table 3).

Table 3 : Variation in serum Lp(a) concentrations as a function of age

Age (years)	Lp(a) in g/l
≤ 35	0.27±0.16
]35-45]	0.23±0.16
]45-55]	0.37±0.17
> 55	0.40±0.27

Bivariate analysis shows no correlation between Lp(a) and other cardiovascular risk factors as shown in Table 4.

Table 4 : Correlation between lipoprotein (a) and others cardiovascular risk factor

Parameters	Obesity	HBP	*Sexe*	Age
	R=−0.14	R=−0.08	R=−0.08	R=−0.009
Lp(a)	p=0.392	p=0.611	p=0.608	p=0.954

Figure 2 indicates a variation in serum Lp(a) levels depending on the accumulation of cardiovascular risk factors. Serum Lp(a) concentrations appear to be decreased when the number of risk factors increases.

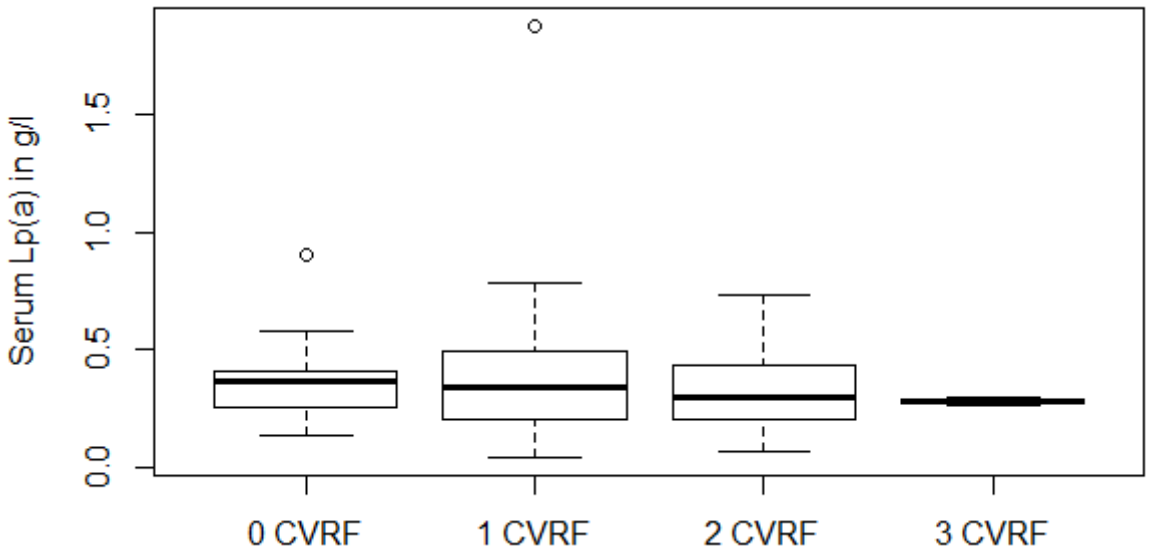

Figure 2 : Variation of serum Lp(a) levels in diabetic subjets depending on the accumulation of cardiovascular risk factors

Discussion

Lp(a) is not systematically measured in routine cardiovascular risk assessment. In Africa and developing countries, few studies have been carried out on this biological marker; in Senegal in particular, we have not found a study that has

been carried out on this subject.Lp(a) has thrombogenic and atherogenic characteristics, and in the blood circulation, it constitutes an antagonist of plasminogen which is the inactive precursor of plasmin, a major enzyme of the fibrinolytic system. It competes with plasminogen at the binding sites of endothelial cells, thus blocking the formation of plasmin, leading to a delay in fibrinolysis and its deposition on the vascular wall [**5, 6**].High levels of Lp(a) therefore lead to a high thrombotic risk due to inhibition of fibrinolytic mechanisms [**6**]. Elevated serum Lp(a) concentrations have long been reported to be associated with increased risk of ischemic cardiovascular disease and, in particular, coronary heart disease [**7**].This leads some authors to consider the measurement of total blood Lp(a) as a screening tool and could prove to be a useful biomarker for detecting patients at high risk of cardiovascular disease [**8, 9**]. The prevalence of hyperlipoproteinemia(a) can be up to 20% in the general population [**7, 10, 11**].These results are superimposable to those of our present study since we obtained a prevalence of hyperlipoproteinemia (a) of 17.8% in healthy subjects. On the other hand, the prevalence is much higher in our type 2 diabetic patients where it is 29.7%. This prevalence of hyperlipoproteinemia (a) in type 2 diabetics varies from one study to another and from one country to another.A Moroccan study by Idrissi et *al* [**12**] showed a prevalence of around 31.2% which is almost identical to ours while a Sudanese study by Mohieldein et al [**13**] presented a prevalence of 51.4% which is significantly higher than our results. These differences observed in the prevalence of hyperlipoproteinemia (a) within these different studies could be linked to the ethnic and racial variability of the populations studied,to the different dosing methods used but also and above all to the usual values taken into consideration as a threshold value for the risk of cardiovascular disease.In our study population, the mean values of the LDL/HDL ratio and the mean serum Lp(a) levels were significantly higher in type 2 diabetic subjects than in controls. Our findings were confirmed by several other studies [**14, 15**]. However, the Sudanese study

by Mohieldein et *al* [**13**] showed results which contradict ours where the mean serum Lp(a) values were significantly higher in their healthy controls compared to their type 2 diabetic patients.Plasma Lp(a) concentrations are generally established by age 5 and remain constant throughout adulthood [**11**] but the pathophysiological mechanism by which individuals with type 2 diabetes have higher plasma levels has not been well elucidated, Hernández et al [**16**] believe that triglycerides and urinary albumin excretion rate are the main factors influencing serum Lp(a) levels in the diabetic population. In the type 2 diabetic population, serum lipoprotein(a) concentrations are higher in women than in male patients [**17, 18**], which corroborates our results.Plasma concentrations of Lp(a) differ according to sex, in men as in women, their increase is linked to advanced age (aging) and in women more particularly to estrogenic status because Lp(a) levels) of postmenopausal women are higher than those of premenopausal women [**19, 20**].This increase in serum Lp(a) levels in relation to age is confirmed by our study where we observed an increase in the average values of our patients over the age of 45. Furthermore, this study found no relationship between Lp(a) and other risk factors proving this marker as an independent risk factor and this was confirmed by Laraqui A et *al* [**21**].Our results revealed a decrease in serum Lp(a) concentrations when the number of cardiovascular risk factors increased.We have not found studies in the literature confirming or refuting these observations, hence the need to conduct longitudinal and cross-sectional studies that could elucidate this enigma. Racial variability has especially been noted with regard to plasma Lp(a) concentrations, with individuals of African ancestry having on average higher concentrations than individuals of European or Asian ancestry [**7, 22, 23**].These important remarks regarding the average plasma values of Lp(a) in black individuals deserve to be taken into consideration with the carrying out in Senegal of independent studies with the aim of establishing the reference values of Lp(a)and to be able to correctly manage subjects with hyperlipoproteinemia (a).

Conclusion

This study shows a high frequency of hyperlipoproteinemia (a) in type 2 diabetics. Female patients have higher plasma levels than male individuals.Serum Lp(a) concentrations are not correlated with other cardiovascular risk factors and therefore constitute an independent risk factor.In Senegal, serumLp(a) measurementisrarely or not carried out, itisthereforeimperative to establishreference values for thisbiologicalparameter in order to set the decisionthreshold for hyperlipoproteinemia (a) and to be able to correctly manage type 2 diabetics.

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