

Lipid profile in type 2 diabetic subjects aged 40 years and over living in Benin

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Abstract

Background: Abnormal lipid profile is common in subjects with type 2 diabetes (T2D). Despite use of lipid-lowering agents, many subjects with T2D do not achieve lipid targets. The present work aimed to assess lipid profile in a randomly selected group of adult diabetic subjects under treatment in order to determine how diabetes treatment affects blood lipid parameters levels. HbA1c level is used as a biomarker of glycaemia control achievement and treatment success in diabetic.

Methods: This study was carried out as a pre-post test design study with a control group. A total of 117 diabetic subjects under treatment and 100 non-diabetics as control subjects are included in the study. TC, TG and HDL-C were measured by enzymatic methods and LDL-C was determined by Friedewald formula. Plasma glucose was measured by Glucose Oxidase method and glycosylated hemoglobin (HbA1c) with a radioimmunoassay.

Results: The results showed that TC ($p < 0.001$), LDLc ($p < 0.001$) and TG ($p = 0.02$) levels increased significantly in diabetics compared to controls. Atherogenic indices TC/HDL-C ($p < 0.01$) and LDL-C/HDL-C ($p < 0.01$), and blood calcium level ($p < 0.001$) were significantly increased in diabetics than in controls. Subjects with HbA1c value $> 7.0\%$ had significantly higher levels of TC ($P < 0.01$), LDL-C ($p < 0.05$), TC/HDL-C ($p < 0.01$), LDL-C/HDL-C ($p = 0.02$), TG/HDL-C ($p = 0.01$) and calcemia ($p < 0.001$) compared to subjects with HbA1c $\leq 7.0\%$.

Conclusion: Significant difference of lipid parameters was observed in diabetics with HbA1c $\leq 7.0\%$ and $> 7.0\%$. Abnormal lipid patterns and insulin resistance tend to be normalized with therapy of diabetes.

Keywords: Type 2 diabetes, HbA1c, lipid parameter, insulin resistance, Benin

1. Introduction

Type 2 Diabetes (T2D) is a hereditary, chronic and endocrine metabolic disorder which causing deaths worldwide [1]. Its prevalence is on rising in developing countries [2,3]. Due to genetic disposition and life style, certain ethnic and racial groups, especially in Africa and Asia, have a greater risk of developing diabetes [4]. The chronic hyperglycaemia of diabetes is associated with long-term damage, dysfunction, and failure of various organs, such as the eyes, kidneys, nerves, heart, and blood vessels [5]. Glycosylated hemoglobin (HbA1c) is a routinely used biomarker for long-term glycemic control. Diabetes treatment is adjusted based on the HbA1c results, expressed as the percentage of hemoglobin that is glycosylated. A vast majority of assays have been standardized worldwide through the

National Glyco-hemoglobin Standardization Program [6] that established the relationship between HbA1c levels and risk for long-term diabetes complications. Nigeria, the most populous country in Africa, is reported to harbour a substantial number of individuals living with diabetes [7]. Abnormal lipid profile is reported tightly associated with diabetes complications in Nigeria [8]. In Benin, which is a close country of Nigeria, little is known about lipid and diabetes association in diabetes suffering population.

Abnormal lipid profile is more common in diabetes and is aggravated with a poor glycaemia control. Then the determination of lipid profile in diabetic subjects is required to investigate how lipid metabolism is affected by diabetes per se and its

treatment. The present work aimed to assess lipid profile in a randomly selected group of adult diabetic subjects under treatment in order to determine how diabetes per se and its treatment affect blood lipid parameters levels. HbA1c level is used as a biomarker of glycaemia control achievement and treatment success in diabetic subjects.

2. Material and methods

2.1 Study participants

This study was carried out as a pre-post test design study with a control group at *Hôpital Saint-Jean* (Cotonou, Benin). From March 2014 to August 2015, a total of 117 subjects (49 females and 68 males) suffering from diabetic and visiting the hospital were enrolled in the study. All diabetic subjects were under treatment and were aged 40 years and over. Diabetic subjects aged less than 40 were excluded from the study. One hundred (100) age- and sex-matched healthy subjects with no complaints and no known diseases were randomly selected at the same hospital and enrolled as control group. Laboratory tests were used to confirm the absence of diabetes in the control group and also by asking questions about signs of diabetes such as polyuria, polydipsia and recent weight loss. Informed written consent was voluntarily obtained from each participant before entering the study and the local research ethics board approved the study protocol.

2.2 Measurement of Biochemical Parameters

Venous blood samples were collected from all the individuals after at least 8 hours fasting. The samples were centrifuged and serum fasting blood glucose concentrations were measured within 30min to 1h following blood samples collection. Plasma glucose was measured by Glucose Oxidase and Peroxidase (GOD-POD) method (ELITech Group, Puteaux, France) according to the manufacturer's instructions. Blood samples for HbA1c were obtained at baseline. HbA1c levels were measured with a radioimmunoassay (Roche HbA1c and Roche Tinaquant; Roche Diagnostics, Meylan Cedex, France). TC (ELITech Group, Maizy, France), high density lipoprotein cholesterol (HDL-C) (Biolabo, Maizy, France) and TG (ELITech Group, Maizy, France) were assayed by enzymatic methods. Low density lipoprotein cholesterol (LDL-C) was determined using the Friedewald's formula $LDL-C = TC - (HDL-C + TG/5)$ when TG values were under 400 mg/dl. According to National Cholesterol Education Programme (NCEP) Adult Treatment Panel III (ATP III) guideline [9], hypercholesterolemia is defined as TC >200 mg/dl, high LDL-C when value >100

mg/dl, hyper-triglyceridemia as TG >150 mg/dl and low HDL-C when value <40 mg/dl.

2.3 Statistical analyses

Data were analysed by SigmaPlot statistical analysis software 2010 (Systat Software, Inc. San Jose, CA, USA). Means and standard errors of the mean (SEM) of blood parameters were calculated. Student's t-test was used to ascertain any difference between the group characteristics. A p value of < 0.05 was deemed significant.

3. Results

3.1 Clinical characteristics of the study subjects

Table 1 shows demographic data of subjects and control participants. The mean (SEM) age of the study subjects was 51.40 (8.30) years for controls and 57.05 (6.05) years for diabetic subjects. Ages were ranged from 40–84 years for both groups. Women represent 43% of the controls and 49% of diabetic subjects. The BMI level in control participants ($27.5 \text{ kg/m}^2 \pm 3.7$) was significantly different from the level in diabetic subjects ($31.0 \text{ kg/m}^2 \pm 9.1$). Average duration of diabetes was 3 years, ranged from 11 months to 15 years.

Table 1: General characteristic of type 2 diabetic Subjects and in controls

Characteristics	Diabetics (n=117)	Controls (n=100)
Age (years)	57.05 ± 6.05	51.40 ± 8.30
Women (%)	49	43
BMI (kg/m ²)	31.0 ± 9.1	27.5 ± 3.7

3.2 Lipid abnormalities

Biochemical parameter levels are shown in table 2. Blood glucose level was significantly ($p < 0.001$) higher in diabetics ($1.89 \pm 0.09 \text{ g/l}$) compared to controls ($0.80 \pm 0.03 \text{ g/l}$). Like glycaemia, HbA1c level was significantly higher ($p < 0.001$) in diabetic subjects ($9.34 \pm 0.21\%$; ranged from 3.92% to 14.51%) compared to $4.44 \pm 0.10\%$ in controls (ranged from 4.63% to 5.80%). However, no strong correlation between glycaemia and HbA1c in diabetic was observed (Fig. 1). Diabetics have significantly higher TC ($p < 0.001$), TG ($p = 0.02$) and LDL-C levels ($p < 0.001$) compared to non-diabetic. HDL-C level in diabetics was not significantly ($p = 0.86$) different from that in non-diabetic subjects. Among all the lipid parameters, only LDL-C level was beyond reference value in diabetics (124.94 mg/l vs. 100 mg/l). We calculated atherogenic indices in order to determine how these predictors of disease are associated with dyslipidemia in T2D subjects.

TC/HDL-C ($p < 0.01$) and LDL-C/HDL-C ($p < 0.01$) in diabetics were significantly increased compared non-diabetic subjects. TG to HDL-C ratio, which is considered as reliable as fasting serum insulin levels, was determined to assess insulin resistance statute in diabetic subjects [25]. TG/HDL-C ratio was

significantly ($p < 0.001$) higher in diabetic subjects than in controls. Blood calcium level was significantly ($p < 0.001$) higher in diabetics than in controls while magnesium level showed no significant difference ($p = 0.12$).

Table 2: Biochemical parameters in Type 2 Diabetic Subjects and in controls

Parameters	Reference range	DT2 (n=117)	Controls (n=100)	p-Value
Glucose (g/l)	0.65-1.10	1.89 ± 0.09	0.80 ± 0.03	< 0.001
HbA1c (%)	4-6	9.34 ± 0.21	4.44 ± 0.10	< 0.001
TC (mg/dl)	< 200	198.09 ± 46.30	141.52 ± 6.78	< 0.001
Triglycerides (mg/dl)	< 150	112.50 ± 5.95	81.48 ± 8.76	0.02
HDL-C (mg/dl)	> 40	49.58 ± 1.97	48.78 ± 4.60	0.86
LDL-C (mg/dl)	< 100	124.94 ± 5.00	76.44 ± 6.02	< 0.001
TC/HDL-C	< 5	4.63 ± 0.20	3.51 ± 0.35	0.01
LDL-C/HDL-C	< 3.5	3.04 ± 0.16	1.96 ± 0.24	< 0.01
TG/HDL-C	nd	3.11 ± 0.28	1.75 ± 0.15	< 0.001
Calcium (mg/l)	90-105	89.28 ± 3.20	107.58 ± 1.04	< 0.001
Magnesium (mg/l)	16-25	17.83 ± 6.89	19.23 ± 0.28	0.13

nd: non determined

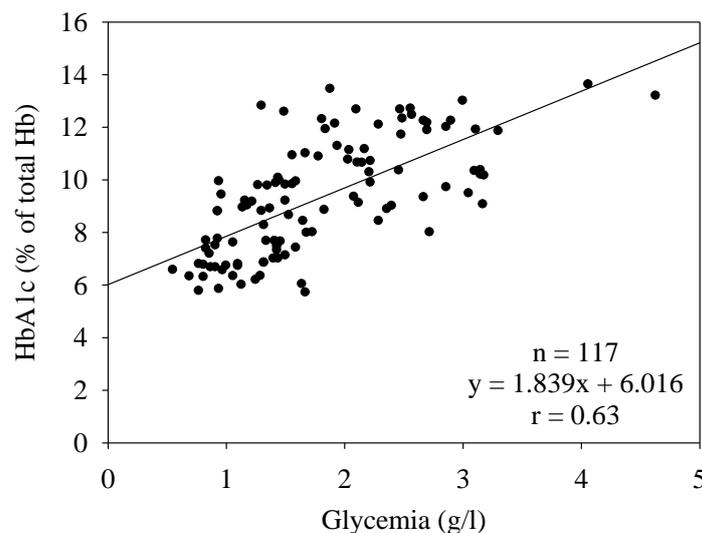


Figure 1: Correlations between HbA1c and glycemia in diabetic subjects.

To determine how glycaemia control achievement affect biochemical parameters, diabetic subjects were classified into 2 groups. First group consists of subjects with HbA1c value $\leq 7.0\%$ and second group consists of subjects with HbA1c value $> 7.0\%$. The results are shown in table 3. Subjects with HbA1c value $> 7.0\%$ had significantly higher value of blood glucose ($p < 0.001$), TC ($P < 0.01$), HDL-C ($p < 0.01$), and LDL-C ($p < 0.05$). TG ($P = 0.17$) level showed no significant changes. Both TC/HDL-C ($p < 0.01$) and LDL-C/HDL-C ($p = 0.02$) levels were

significantly increased in diabetic with HbA1c value $> 7.0\%$ compared to subjects with HbA1c $\leq 7.0\%$. TG/HDL-C level was significantly ($p < 0.01$) higher in case of HbA1c value $> 7.0\%$ compared to when HbA1c $\leq 7.0\%$. Blood calcium level also varied in diabetic population. Calcemia level, which was lower in diabetic subject than in controls, decreased even further in diabetic subjects with HbA1c value $> 7.0\%$ compared to subjects with HbA1c $\leq 7.0\%$ ($p < 0.01$). Blood magnesium level showed no significant changes between both groups of diabetic subjects.

Table 3: Biochemical Parameters categorized by subjects' glycemic controls (HbA1c)

Parameter	Glycated Hemoglobin (HbA1c)		P-value
	≤ 7.0 (n=100)	> 7.0 (n=117)	
	Mean ± SEM	Mean ± SEM	
Glucose (g/l)	1.27 ± 0.19	2.05 ± 0.10	< 0.001
TC (mg/dl)	173.98 ± 11.50	203.03 ± 9.56	0.01
Triglycerides (mg/dl)	97.00 ± 8.95	117.00 ± 7.03	0.17
HDL-C (mg/dl)	51.46 ± 5.40	37.49 ± 2.21	< 0.01
LDL-C (mg/dl)	105.15 ± 10.48	131.14 ± 5.45	< 0.05
TC/HDL-C	3.63 ± 0.28	4.84 ± 0.23	0.01
LDL-C/HDL-C	2.23 ± 0.21	3.63 ± 0.19	0.02
TG/HDL-C	2.80 ± 0.21	4.73 ± 0.30	0.01
Calcium (mg/l)	95.42 ± 6.17	84.22 ± 1.53	0.001
Magnesium (mg/l)	18.53 ± 1.81	16.87 ± 0.37	0.07

4. Discussion

The determination of serum lipid parameters is required in diabetic subjects care. To understand the mechanism underlines the changes in lipoproteins in diabetes mellitus and the way by which this may influence the development of the cardiovascular disease that accompanies this disorder, we examine the metabolism of lipoproteins. Screening and treatment of lipid abnormalities prevents and reduce the risk of cardiovascular events [10-12]. However, lipid profile determination is not a routine exam in diabetic treatment monitoring in Benin. The present work aimed to assess lipid profile in a randomly selected group of adult diabetic subjects under treatment in order to determine how diabetes per se and its treatment affect blood lipid parameters levels. HbA1c level is used as a biomarker of glycaemia control achievement and treatment success in diabetic subjects.

Diabetes is a chronic disease whose complications appear only after several years. By conducting this study in diabetics aged 40 years and over, we hoped to achieve explicit outcomes. We measured the serum levels of TC, TG, HDL-C and LDL-C in diabetics subjects that we compared to those from non-diabetic subjects. A total of 217 subjects participated to the study including 117 with T2D and 100 non diabetic subjects as the control group. The group of diabetic subjects was equally composed of women and men. Diabetic subjects were predominantly overweight (83%). This result is far higher than the average proportion of overweight or obese in West Africans which were estimated at 31.4% [13]. Obesity is an additional risk factor for diabetes [14] and our finding relative to the high rate of overweight in diabetics is of paramount concern.

The results of HbA1c are very revealing. In the control group, the values were from 4.0 to 5.0% and no diabetic had HbA1c result <7.0%. Many

diabetics had blood sugar in the normal range and the correlation between blood glucose and HbA1c in diabetics was of poor quality. This suggests that blood sugar is not a good biomarker in diabetics. HbA1c level has been suggested to be used for the diagnosis of diabetes [1]. Our results strongly support this idea. For this purpose, Mayer and Schriger [15] reported that whether an individual has an initial HbA1c level of $\geq 7.0\%$ but on a repeat test has a level $< 7.0\%$, the diagnosis should be pre-diabetes. These individuals should be treated appropriately and periodically retested to ensure that their HbA1c level remains below 7.0%.

It is known that subjects with diabetes can have many complications including elevated levels of VLDL-C, LDL-C and TG, and low levels of HDL-C [16]. In the present study, the results showed that lipid parameters TC, TG, LDL-C levels of the diabetics were higher than those in the controls. Our results are in agreement with other findings [8,17] but with somewhat less profound lipid profile damage. This difference could be explained by the fact that diabetics in our sample were under treatment. Among all the lipid parameters, only LDL-C level was beyond reference value in diabetics. Elevated LDL-C is a major risk factor for cardiovascular disease [18]. As such, management of LDL-C is the primary goal of therapy for diabetic dyslipidemia [19,20].

Atherosclerosis and cardiovascular disease are common in clinical cases where dyslipidemia is present. We estimated the atherogenic indices TC/HDL-C and LDL-C/HDL-C in order to determine whether diabetics were at risks of cardiovascular diseases. Our results showed that the atherogenic indices were higher in diabetics than in controls. However, these levels were within the normal ranges [9] suggesting that diabetics in our study were at low risk of cardiovascular disease and that the fundamental of their treatment is good. Our finding that blood Ca^{2+} level is low in diabetics compared to

controls is in agreement with the report that this ion plays important role in insulin secretion [21], and that insulin level must be decreased in diabetics [22].

HbA1c was established as the gold standard of glycemic control. The level of HbA1c value $\leq 7.0\%$ was said to be appropriate for reducing the risk of cardiovascular complications [23]. In the present study, we determined that diabetics with HbA1c value $>7.0\%$ had worsen lipid profile compared to diabetics with HbA1c $\leq 7.0\%$. As elevated HbA1c and dyslipidemia are independent risk factors of cardiovascular disease, diabetic subjects with elevated HbA1c and dyslipidemia can be considered at higher risk group for cardiovascular disease [24]. Our finding that both atherogenic indices TC/HDL-C and LDL-C/HDL-C were significantly increased and beyond references values is in accordance with the notion that bad glycemia control augment complication and cardiovascular disease risks in diabetics [23,24]. It was reported that a simple TG/HDL-C ratio is a good marker of insulin resistance [25]. Insulin resistance is the basic pathology underlying type 2 diabetes. Here, we showed that TG/HDL-C level was increased in diabetic than in controls and in diabetic with HbA1c $>7.0\%$ compared to diabetics with HbA1c $\leq 7.0\%$. Our results support the notion that TG/HDL-C ratio can be used as a surrogate of insulin resistance [25] and that HbA1c level control under treatment reflects insulin resistance improvement.

5. Conclusion

Plasma TC and TG are significantly elevated in diabetics compared to non-diabetics. Significant difference of lipid parameters was observed in diabetics with HbA1c $\leq 7.0\%$ and $>7.0\%$ confirming that HbA1c can be used as a potential biomarker for predicting dyslipidemia in T2D subjects. Abnormal lipid patterns and insulin resistance tend to be normalized with therapy of diabetes.

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