

Evaluation of High Density Lipoproteins Characteristics in Type 2 Diabetics

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ABSTRACT

To investigate the characteristic features of high density lipoproteins in type 2 diabetes mellitus patients, we examined the lipid parameters, quantitated glucose and insulin levels along with the apolipoprotein ratios. Using radial immunodiffusion, colorimetric, flourimetric and ultracentrifugation methods the values of the parameters were obtained. We observed a moderate elevation of the Hdl/Ldl cholesterol and apolipoprotein A-1/A-11 ratio which are exponents risk factors in the exacerbation of atherosclerosis. Both males and females show higher level of these parameters when compared with controls and were markedly higher ($p > 0.05$). We also noticed a marked elevated correlation ($r = 0.891$, $p < 0.01$) between the high density lipoprotein and the apoprotein A-1/A-11 ratio. Understanding that Hdl of diabetic patients are enriched with triglyceride and cholesterol depleted we reason that changes that may occur as a result of poor management of diabetes has the ability to decrease the antiatherogenic effect of high density lipoprotein. We could advance evidence that an elevated apolipoprotein A-1/A-11 ratio in type 2 diabetic patients has the capacity to inhibit the development of atherosclerosis.

Keywords: High density lipoprotein, lipids, characteristics, type 2 diabetes

INTRODUCTION

For lipids to perform their metabolic functions effectively in tissues and organs, they must be transported. This transport is effectuated by micelles called lipoproteins. Atherosclerosis which is a complication of diabetes has elicited major health concern. A vascular disease accounting for over 70% of diabetic mortality and morbidity, it is now known to be a complication of type 2 diabetes mellitus. Previous studies gleaned from periodicals opines that the incidence and prevalence of atherosclerosis are inversely correlated with the level of plasma high density lipoprotein (Hdl) ^[1,20]. It is yet not clear if a similar scenario is presented for type 1 diabetes. The triad of atherosclerosis, type 2 diabetes mellitus and cardiovascular disease requires further consideration in developing therapies. High density lipoprotein (Hdl) plays diverse roles in cholesterol transport, exuding vascular-protection, and elucidating endothelium related vaso-relaxation, antioxidation, anti-inflammatory as well as anti-thrombotic functions ^[2,24]. There is now enough evidence to input that a strong relationship exist between the severity of type 2 diabetes and an enervated Hdl and apoprotein A-1 and A-11 functionality ^[3]. Studies has shown that in combination with Hdl cooperativity exist with sphingosine-1-phosphate (SIP) to express their biological function ^[4]. SIP is known to be a facilitator of the protective effect of Hdl including the ability to inhibit reverse atherosclerosis. Studies have pointed in a direction which elucidated the fact that atherosclerosis is a multifactorial inflammatory disease in which prostanoids and cyclooxygenase play diverse roles to enhance the disease process ^[5]. It is now known that SIP combines with Hdl exhibiting synergistic function in induction of the Cox-2 expression and prostaglandin 1-2 release ^[6].

There is evidence to show that in type 2 DM, the capacity of the Hdl playing a protective function is impaired ^[7,22,23]. The need to re-evaluate the biochemical components of Hdl and other lipids in terms of structure and function using more advance techniques like nuclear magnetic resonance to increase understanding is encouraged.

MATERIALS AND METHOD

At the Federal Medical centre, Yenagoa, Bayelsa State, Nigeria, blood (serum, plasma) and urine samples were collected from type 2 diabetic patients ($n=60$), from (males=30) and (females=30). Blood glucose levels of subjects were above threshold value ≥ 10 mmol/l with presence of glucosuria and ketonuria. Control subjects for the experiment were those with normal glucose levels ranging between 3.5-6.7 mmol/l with no glucosuria and ketone in urine. All samples were over night fasting samples.

ANALYTICAL METHODS

The concentration of fasting blood glucose was determined by the glucose oxidase method with the use of Randox Kits (London) and values measured with a spectrophotometer SPEC22D⁺ set at 540nm. Urine samples were analysed with N-multistix Macherry-Nagel (Germany) for glucose and ketones.

Triglyceride and cholesterol were determined colorimetrically with Corning colorimeter. Cholesterol was measured by a Lieberman Burchard reagent method. The triglyceride were determined by a fluorimetric 24 pentanedione procedure calibrated with inolein.

HDL cholesterol was determined by measurement of cholesterol in the supernatant after precipitation of VLDL and LDL in 2.0ml of plasma with 0.15ml of 1M MnCl₂ and 0.12ml of sodium Heparin (35mg/ml, Ricker) [19, 8,21]. The LDL cholesterol was derived from the Friedewald equations as follows: LDLcholesterol = total cholesterol-(HLD Cholesterol+Triglyceride/5).

Plasma Apo A-1 and A-11 were determined by radial immunodiffusion assay.

Serum insulin levels were measured using electrochemical luminescence immunoassay (Poch Diagnostics, Manhein Germany).

RESULTS

Mean values of the various parameters estimated for males and females are shown in table 1 and 2 respectively. The data (mean \pm SD) are presented here for all 60 subjects and 60 normal (control). The data show that there was not much variation in the lipoprotein subclass in comparing the two groups. There was however a slight increase in A-1 in female than in males. We also noted that the degree of hyperglycemia also reflects the lipid levels.

Table 1: Apolipoprotein and other lipids levels in Type 2 Diabetic Patients (Males)

Parameters	Control (n=30)	Diabetics (n=30)	p-value ≥ 0.05
FBG (mmol/l)	4.8 \pm 1.5	15.8 \pm 3.0	
Insulin (Pmol/l)	125 \pm 5.0	145 \pm 3.10	
Triglyceride (mmol/l)	1.5 \pm 0.3	1.7 \pm 0.3	
Cholesterol (mmol/l)	4.3 \pm 1.5	6.8 \pm 0.8	
LDL Chol. (mmol/l)	3.35 \pm 1.2	5.7 \pm 3.06	
HDL chol. (mmol/l)	0.40 \pm 0.2	0.6 \pm 0.22	
HDLC/LDLC	0.12 \pm 0.01	0.10 \pm 0.01	
APO A-1 (g/l)	1.53 \pm 0.8	1.8 \pm 0.5	
APO A-11(g/l)	0.52 \pm 0.05	0.7 \pm 0.03	
A-1/A-11(g/l)	2.8 \pm 0.2	3.2 \pm 0.03	
Urine glucose	Nil	++	
Urine ketones	Nil	++	

Values mean \pm SD

Table 2: Apolipoprotein and other lipids level in type 2 Diabetic Patients (Females)

Parameters	Control (n=30)	Diabetics (n=30)	p-value ≥ 0.05
FBG (mmol/l)	4.6 \pm 1.4	14.8 \pm 4	
Insulin (pmol/l)	124 \pm 6	140 \pm 3.2	
Triglyceride (mmol/l)	1.4 \pm 0.2	1.6 \pm 0.2	
Cholesterol (mmol/l)	4.5 \pm 1.6	6.7 \pm 0.4	
LDL Chol. (mmol/l)	3.4 \pm 1.3	5.6 \pm 3.04	
HDL chol. (mmol/l)	0.4 \pm 0.3	0.52 \pm 0.6	
HDLC/LDLC	0.11 \pm 0.01	0.09 \pm 0.01	
APO A-1 (g/l)	1.82 \pm 0.7	2.0 \pm 0.5	
APO A-11(g/l)	0.50 \pm 0.04	0.8 \pm 0.03	
A-1/A-11(g/l)	3.0 \pm 0.3	3.6 \pm 0.04	
Urine sugar	Nil	++	
Urine ketones	Nil	++	

Values are mean \pm SD

DISCUSSION

We have assessed the roles high density lipoprotein can play in type 2 diabetes mellitus. Using parameters such as glucose, insulin, lipids and the apolipoproteins, the present data suggest that hyperglycemia can be a predisposing factor modulating high density lipoprotein characteristics. The value of HDL/LDL cholesterol and apolipoprotein A-1/A-11 were higher in both males and females when compared with controls. Evidence shown by [8] elucidates this fact. The value of Apo A-1 was moderately higher for females as shown by this study. The difference in the Apo A-1/A-11 ratio for the diabetic patients and controls confer greater understanding. As shown by [6,9] there is significant correlation between the Apo A-1/A-11 ratio and the amount of high density lipoprotein. Among the subclass of Hdl, Hdl₂, has been used to provide indirect evidence for reduced risk of

atherosclerosis. One can adduce possible biochemical mechanism which may be responsible for changes in high density lipoprotein particles. Insulin is known to modify APOA-11 release^[10,18] and in condition of deficiency caused by type 2 diabetes, APOA-11 metabolism can be effected. It has been shown that insulin resistance has effect on lipoprotein subclass production and distribution. Evidence elucidates the fact that elevated insulin sensitivity has the potential to increase mean particle size of VLDL and reduce LDL and HDL particles. Previous studies have also shown that there is a shift in production of LDL and HDL in favour of LDL although a modest increase in small HDL is noticed^[11,12]. Lipoprotein production in favour of Ldl and as it occurs in insulin resistance is a major predisposing factor in arteriosclerosis. It would appear that the lipoprotein subclass changes that occur in type 2 diabetes can be as a resistance. It is now known that HDL is a major carrier of sphingosine-1-phosphate which control major biological activities^[13,14,15]. In diabetes HDL acting in tandem with SIP has the potential to improve vascular system function^[16,17]. Additionally SIP modulates the production of cyclooxygenase.

We have demonstrated that basically all major lipoprotein classes are affected as a function of insulin sensitivity in type 2 diabetes mellitus. Development of methods that will quantitate all subclasses of HDL will improve understanding and promote therapies that will enhance management of diabetes.

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REFERENCES

- (1). Beach, K.W., Bruzell, J.D., Conquest, L.L., Strandress, D.E., (1979). The Correlation of arteriosclerosis obliterans with lipoproteins in insulin-dependant and non-insulin dependant diabetes. *Diabetes* 28:836-40.
- (2). Maya, S., Gowri, Deneys, R., Van der Westhuyzen, Susan, R., Bridges, James, W., Anderson (1999). Decreased protection by HDL from poorly controlled type 2 diabetic subjects Against LDL oxidation may be due to the abnormal composition of HDL. *Artherosclerosis Thrombosis and Vascular Biology* 19:2226-2233.
- (3). Anotol, A., Kontush and Chapman, M. J. (2006). Functionality defective High-density-lipoprotein: A new therapeutic target at the crossroad of dyslipidemia, inflamatroc and Atherosclerosis. *Pharmacol Rev.* 58:342-374.
- (4). Christopherson, C., Obinata, H., Kumazaswamy, SB, Galvani, S., Alinstrom, I., Servana, M., Egerar-Sieber, C, Muller, VA, Hla, T., Nielsen, L.B., (2011). Endothelium-protective sphingosine-1-phosphate provided by HDL-associated apolipoprotein M. *Proc. Natl acad sci USA* 108(23):9613-9618.
- (5). Liu, D., Ji, L., Wang, Y., Zheng, L., (2012). Cyclooxygenase=2 Expression, prostacyclin production and Endothelial protection of high-density lipoprotein. *Cardiovax. Hematol Disord Drug Targets* 12:98.
- (6). Samia, M., James, D.O., Robert, S.R., Aruna, P., Julie, E.B. and Paul, M.R. (2010). Lipoprotein particle size and concentration by nuclear magnetic resonance and incident type 2 diabetes in women. *Diabetes* 59:5. 1153-1160.
- (7). Neeli, H., Gadi, R., Rader, D.J, (2009). Managing diabetic dyslipidemia: beyond statin therapy. *Curr Diab Rep.* 9(1):11-17
- (8). Tomkin, G.H., Owens, D., (1994). Insulin and lipoprotein metabolism with special reference to the diabetic state. *Diabetes Metab Rev* 10:225-252.
- (9). McNamara, J.R., Jeaner, J.L., Li, Z., Nilson, W.F., Schaefer, E.J., (1992). Change in LDL: Particle size in associated with plasma triglycerides concentration. *Artheroscler Throm* 12:1284-1290.
- (10). Defronzo RA, Ferrannin, E. (1991) Insulin resistance. A multifaceted syndrome responsible for NIDDM, obesity, hypertension dyslipidemia and atherosatirotic cardiovascular disease. *Diabetes care.* 14:173-194.
- (11). Martin, H.K. (2004). Evaluating interference caused by lipemia. *Clinical Chemistry.* 50:11. 1968-1969.
- (12). Ronald, M., Krauss, M.D. (2004). Lipids and lipoprotein in patients with type 2 diabetes. *Diabetes Care.* June 1, 27 (6) 1496-1504.
- (13). Lee, M.H., Hammad, S.M., Semler, A.J., Lutrel, L.M., Lopes-Virella, M.F., Klein, R.L. (2010). HDL, but not HDL₂, stimulates plasmogen activator inhibitor-1-release from adipocytes: the role of sphingosine-1-phosphate. *J. Lipid Res* 51(9) 2619-2628.
- (14). Friedlander, Y., Kidron, M., Caslake, M., Lamb Tracey, McConell, Bar-on H (2000). Low density lipoprotein particle size and risk factors of insulin resistance syndrome. *Artherosclerosis.* 148:141-149.
- (15). Plutzky, J., Viberti, G., Hoffner, S. (2002). Artherosclerosis in type 2 diabetes mellitus and insulin resistance: mechanistic links and therapeutic targets. *J. Diabetes Complications,* 16(6):401-415.

- (16). Eren, E., Yimaz, N, Ayin, O. (2012). High density lipoprotein and its dysfunction. *Open Biochem J.* 6:78-93.
- (17). Otvos, J.D. (2002). Measurement of lipoprotein subclass profiles by nuclear magnetic resonance spectroscopy. *Clin Lab* 48:171-180.
- (18). Teskinen, M.R. (1995) Insulin resistance and lipoprotein metabolism. *Curr Opin lipidol* 6:153-160.
- (19). Chung, B.H., Williamson, T., Geer, J.C., Segres, J.P., (1980). Preparative and quantitative Isolation of plasma lipoprotein: rapid, single discontinuous density gradient ultracentrifugation in a vertical rotor, *J Lipid Res* 21(3): 234-241.
- (20). Mooradeari, A.D, (2009). Dyslipidaemia in type 2 diabetes mellitus. *Nat Clin Prac Endocrinal Metab* 5(3):150-159.
- (21). Patsch, W., Schofeld, G., Gotto, A.M. Jr., Patsch, J.R. (1980). Characterization of human high density lipoproteins by zonal ultracentrifugation. *J. Biol Chem* 255:3178-85.
- (22). Mario, C., de Beer, Nancy, R.W., Joanne, M.W., Victoria, P.N., Debra, L.R., Ailing, J.I., Deneys, R.V. (2010). Impact of serum amyloid A on high density lipoprotein composition and levels. *J. Lipids Res.* 51:3117-3125.
- (23). Sorci-Thomas, M.G., Thomas, M.J, (2012). High density lipoprotein biogenesis, cholesterol efflux, and immune cell function, *Artheroscler Throm Vasc Biol* 32(11):2561-2565.
- (24). Liu, D., Ji, L., Zhan, D., Tong, X., Pan, B., Liu, P., Zhang, Y., Huang, Y., Su, J., Willad, B., (2012). Nonenzymatic glycation of high density lipoprotein impairs its anti-inflammatory effects in imate immunity. *Diabetes Metab Res Rev* 28(2):186-195.

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