International Journal of Health Sciences and Research
www.ijhsr.org ISSN: 2249-9571

Original Research Article

Glycosylated Hemoglobin (HbA_{1C}): Association with Dyslipidemia and Predictor of Cardiovascular Diseases in Type 2 Diabetes Mellitus Patients

Abdulrahaman A Momin[1*], Mangesh P Bankar[2], Gouri M Bhoite[3]

[1]Assistant Professor, Department of Biochemistry, Institute of Medical Sciences & Research, Mayani, Satara, Maharashtra, India
[2]Professor, Department of Biochemistry, B. J. Government Medical College, Pune, Maharashtra, India
[3]Assistant Professor, Bharati Vidyapeeth University Dental College, Pune, Maharashtra, India

[*]Correspondence Email: rahaman.momin@gmail.com

Received: 06/05//2013 *Revised: 11/06/2013* *Accepted: 17/06/2013*

ABSTRACT

Diabetes is a metabolic disorder characterized by hyperglycemia, either due to insulin deficiency or insulin resistance. Despite some progress in the development of new anti-diabetic agents, the ability to maintain tight glycemic control in order to prevent complications of diabetes without adverse complications still remains a challenge. In present study100 type 2 diabetic patients and 100 age and sex matched control subjects were included. We investigated the blood glucose, lipoproteins, atherogenic ratios and glycosylated haemoglobin in study and control group. Further the type 2 diabetic subjects were divided in to two groups depending on glycemic index, first group consists of patients with HbA1c level ≤7.0% and second group consists of patients with HbA1c level >7.0%. We found significant increase in the levels of total serum cholesterol, triglyceride, LDL cholesterol and VLDL cholesterol in patients with type 2 diabetes than control subjects. Also the atherogenic ratios viz. total cholesterol/HDL cholesterol and HDL cholesterol/LDL cholesterol differ significantly differ in type 2 diabetic subjects as compared to controls. The HDL cholesterol was significantly lower in type 2 diabetics. The patients with poor glycemic control (HbA1c>7.0%) had significantly higher levels of total cholesterol, LDL cholesterol, total cholesterol/HDL cholesterol ratio and decreased HDL cholesterol/LDL cholesterol ratio as compared to the patients with good glycemic control (HbA1≤7.0%). These findings suggest that type 2 diabetes patients with dyslipidemia are at increased risk of cardiovascular diseases. The association between HbA1c with various lipid parameters and atherogenic ratios suggest the importance of glycemic control in order to control dyslipidemia and future risk of cardiovascular disease risk in type 2 diabetic patients.

Keywords: Lipid profile, type 2 diabetes mellitus, Glycosylated haemoglobin, Triglyceride, total cholesterol

INTRODUCTION

The worldwide increase in the prevalence of type 2 diabetes mellitus is posing a massive health problem in both developed and developing countries. It is calculated that worldwide there are more than 150 million people with diabetes and

this number will rise to 300 million by 2025. [1]

Type 2 diabetes mellitus is a heterogeneous condition characterized by the presence of both impaired insulin secretion and insulin resistance. Type 2 diabetes mellitus makes up about 90% of the diabetic population. It is chronic disease and usually irreversible. Therefore, for the remainder of their lives patients with diabetes have to consult health care providers on a regular basis. They are prone to certain complications. Glycemic control, control of blood pressure and lipid levels are beneficiary in the prevention or delay in onset & severity of diabetes complications. [2-4]

The chronic hyperglycemia of diabetes is associated with long-term damage, dysfunction and failure of various organs, especially the eyes, kidneys, nerves, heart and blood vessels. [3-5] Type 2 diabetes mellitus is associated with the development of premature arteriosclerosis and a higher cardiovascular morbidity and mortality. Diabetic hyperlipidemia is believed to play an important role in the pathogenesis of accelerated atherosclerosis. [4,6] Silent myocardial ischemia has a reported prevalence of 10-20% in diabetic population as compared to 1-4% in non-diabetic population. [7]

According to NCEP-ATP III guideline,[8] hypercholesterolemia is defined as total cholesterol > 200 mg/dl, high LDL cholesterol when value > 100 m g / dl, hypertriglyceridemia as triglyceride > 150 mg/dl and low HDL cholesterol when value < 40 mg/dl. Dyslipidemia was defined by presence of one or more than one abnormal serum lipid concentration from above.

Characteristic abnormalities in lipids in type 2 diabetes mellitus include elevated triglycerides levels, decreased athero-protective high density lipoprotein cholesterol levels and increased levels of small dense LDL-cholesterol. [3] Dyslipidemia and hyperlipidemia generally coexist in diabetic patients with poor glucose control. Although they have been shown to be independent significant risk factors for vascular complications, the interaction of hyperglycemia and dyslipidemia increases the risk of macro and micro-vascular complications together. Dyslipidemia which includes both quantitative and qualitative abnormalities of lipoproteins can also play significant role in the pathogenesis of vascular complication in patients with type 2 diabetes. [1]

Patients with diabetes can have a reasonably normal life style if they are well educated and motivated regarding the disease. However, most of type 2 diabetic patients belongs to low or middle socioeconomic class and are less educated about the disease. [2] The present study was conducted with the objective to study the lipid profile of type 2 diabetic patients, and to see the effect of glycemic control on lipid profile as a predictor of coronary heart disease in type 2 diabetes mellitus with dyslipidemia.

MATERIALS AND METHODS

This study was conducted in department of biochemistry of B. J. Medical College, Pune. One hundred patients of diabetes mellitus in the period of one year were randomly selected from the outdoor and indoor patient department of Medicine, B. J. Government Medical College, Pune. The age group selected was 40-60 years irrespective of sex.

Patients, who were pregnant, had angina or heart failure, renal failure, hypertension and severe concurrent illness were excluded from the study. Healthy volunteers without any clinical & biochemical evidence of diabetes or hyperlipidemia, chronic kidney or liver

diseases or hyperthyroidism was selected to provide control.

The relevant clinical examination and all routine investigations were performed. An informed consent was taken from every patient after full explanation of procedure. Every patient was advised for at least 12-14 hours overnight fasting and the 2 ml of blood was collected in fluoride bulb for glucose estimation, 2 ml of blood was collected in EDTA bulb for the estimation of glycosylated hemoglobin and 3 ml in plain bulb and serum was separated. Serum samples were analysed for total cholesterol, triglycerides, HDL cholesterol using commercially available kits.

Blood glucose was quantified by GOD-POD method.[9] Glycosylated hemoglobin was estimated by Boronate affinity assay.[10] Serum Total Cholesterol was estimated by enzymatic method. [11] For HDL cholesterol estimation, Chylomicrons, VLDL (very low density lipoproteins) and LDL fractions of cholesterol in serum are separated from HDL by precipitating with phosphotungstic acid and magnesium chloride. After centrifugation the cholesterol in the HDL fraction, which remains in the supernatant is assayed with enzymatic cholesterol method, using cholesterol esterase, cholesterol oxidase, peroxidase and chromogen 4-amino-antipyrine/phenol.[12] Serum Triglyceride was determined by Glycerol phosphate oxidase-peroxidase method.[13] LDL Cholesterol and VLDL cholesterol levels were calculated using Friedwald's formula. [14]

Statistical analysis:

All values were expressed as mean ± standard deviation of mean. Statistical analysis was done by independent samples t-test. The results were significant if $p<0.05$.

RESULTS

Total 100 type 2 diabetic individuals included in this study along with 100 age and sex matched control subjects. The mean duration of diabetes in patients was 4.32 ± 1.39 years. The mean BMI was 26.12 ± 4.06 kg/m^2 in patients and 22.00 ± 3.7 kg/m^2 in controls. WHR was 1.0 ± 0.07 in patients and 0.95 ± 0.085 in the control group (table 1). The levels of BMI and WHR were found to be significant ($P<0.0001$).

Table 1- Mean values of demographic & lipid parameters in type 2 diabetic patients and controls.

Parameters	Controls (N=100)	Patients (N=100)	'P' Value
BMI (kg/m2)	22.00 ± 3.7	26.12 ± 4.06	P < 0.0001
WHR	0.95 ± 0.085	1.0 ± 0.07	P < 0.0001
Duration of diabetes (yrs)	—	4.32 ± 1.39	—
Fasting Blood Glucose (mg/dl)	98.23 ±16.0	246.46 ± 85.26	P < 0.0001
Glycosylated hemoglobin (%)	4.80 ± 1.23	7.79 ± 1.76	P < 0.0001
Total cholesterol (mg/dl)	182.60 ± 33.08	224.81 ± 40.17	P < 0.0001
Triglycerides (mg/dl)	144.30 ± 42.5	185.27 ± 66.52	P < 0.0001
HDL cholesterol (mg/dl)	41.46 ± 4.26	35.60 ± 3.91	P < 0.0001
LDL cholesterol (mg/dl)	112.20 ± 30.73	153.91 ± 39.91	P < 0.0001
VLDL cholesterol (mg/dl)	28.87 ± 8.50	13.30 ± 37.05	P < 0.0001
Total cholesterol/HDL cholesterol	6.35 ± 1.62	4.44 ± 0.94	P < 0.0001
HDL cholesterol/LDL cholesterol	0.40 ± 0.11	0.27 ± 0.09	P < 0.0001

The mean fasting blood glucose was 246.46 ± 85.26 mg/dL for patients and 98.23 ±16.0 mg/dL for controls. The mean values of glycosylated hemoglobin were 7.90 ± 1.9% in diabetics and 4.80 ± 1.23% in controls. The mean values of HbA1c and fasting blood glucose were significantly ($p<0.0001$) higher in patients in comparison to control subjects.

Among the subjects studied in study group reported that, the fasting blood glucose and glycosylated hemoglobin were

positively related with correlation coefficient 0.02, together with the P-value and the 95% confidence interval of P=0.8060 and 0.17 to 0.22 respectively.

The difference between the sample means of serum cholesterol and triglyceride in diabetics and non-diabetics were 42.21 & 40.97 mg/dl, with a 95% confidence interval from 31.28 to 51.82 mg/dl & 40.92 to 78.90 mg/dl; the t test statistic was 7.98 & 6.22, with 198 degrees of freedom and an associated P values of P<0.0001 & P<0.0001 respectively.

The difference between mean levels of LDL (35.12 mg/dl) and VLDL (11.98 mg/dl) cholesterol were found to significant with 95% confidence interval from 24.96 to 45.29 mg/dl & 8.18 to 15.78 mg/dl respectively. The t test value for LDL cholesterol was 6.81 with 198 degrees of freedom and associated P value p<0.0001, and for VLDL cholesterol t value was 6.22, with 198 degrees of freedom and associated P value of P < 0.0001

Importantly, the serum HDL cholesterol level was significantly lower in patients (35.60 ± 3.91 mg/dL) compared to controls (41.46 ± 4.268 mg/dL) with associated P value of P < 0.0001. The range of HDL cholesterol at 95% confidence interval was 6.69 to 4.42, with statistic t value 9.67 and 198 degrees of freedom. As a consequence, the atherogenic ratios of total cholesterol/HDL cholesterol as well as HDL cholesterol/LDL cholesterol were significantly (P<0.0001) higher in type 2 diabetes patients compared to controls with 95% confidence interval 1.54 to 2.28 & 0.16 to 0.10 respectively. The results are shown in table 1

Diabetic patients were classified into 2 groups as per their glycemic index; first group consists of patients with HbA1c value ≤7.0 % and second group consists of patients with HbA1c value >7.0%. Patients with HbA1c value >7.0% had significantly higher value of total cholesterol with associated P value P < 0.0001 and confidence interval 47.37 to 18.34 mg/dl at 98 degrees of freedom, LDL cholesterol with associated P value P = 0.0003 and confidence interval 44.60 to 13.91 mg/dl at 98 degrees of freedom, total cholesterol/HDL cholesterol ratio was significant with P = 0.0002 and HDL cholesterol/LDL cholesterol ratio was significant with P = 0.0011 as compared to the patients with HbA1c value ≤ 7.0%. There was no significant difference found in levels of triglycerides (P=0.1671), HDL cholesterol (P=0.1177) and VLDL (P=0.1671). (Table2)

Table 2- Demographic & lipid parameters categorized by patients glycemic control (HbA1c).

Parameters	Glycosylated Hemoglobin		'P' Values
	≤7.0 (n=43)	>7.0 (n=57)	
BMI (kg/m2)	25.53 ± 0.69	26.59 ± 0.49	P = 0.2014
WHR	1.00 ± 0.01	0.99 ± 0.01	P = 0.3142
Glycosylated hemoglobin (%)	6.12 ± 0.10	9.05 ± 0.16	P < 0.0001
Total cholesterol (mg/dl)	205.37 ± 29.80	238.23 ± 40.35	P < 0.0001
Triglycerides (mg/dl)	190.51 ± 76.78	214.60 ± 91.78	P = 0.1671
HDL cholesterol (mg/dl)	36.67 ± 3.65	35.45 ± 3.96	P = 0.1177
LDL cholesterol (mg/dl)	130.60 ± 32.41	159.86 ± 42.16	P = 0.0003
VLDL cholesterol (mg/dl)	38.10 ± 15.36	42.92 ± 18.36	P = 0.1671
Total cholesterol/HDL cholesterol	5.68 ± 1.18	6.86 ± 1.73	P = 0.0002
HDL cholesterol/LDL cholesterol	0.30 ± 0.11	0.24 ± 0.08	P = 0.0011

DISCUSSION

Lipid abnormalities are common in diabetics and frequently seen in type-2 diabetics. Dyslipidaemias make diabetics prone to develop CHD and other complications of atherosclerosis. Hyperglycemia and atherosclerosis are related in diabetes mellitus. Persistent hyperglycemia causes glycosylation of all proteins especially collagen cross linking and matrix proteins of arterial wall. This eventually causes endothelial cell dysfunction, which is a contributing factor for atherosclerosis. Remnants of triglyceride rich lipoproteins seem to be extremely atherogenic. [1, 6-7]

The actual pathogenesis of diabetic dyslipidemia is not known; many evidences suggest that insulin resistance has a central role in the development of diabetic dyslipidemia. The main cause of diabetic dyslipidemia is the increased free fatty-acid release from insulin-resistant fat cells. [15,16] The increased flux of free fatty acids into the liver in the presence of adequate glycogen stores promotes triglyceride production, which in turn stimulates the secretion of apolipoprotein B and VLDL cholesterol. The impaired ability of insulin to inhibit free fatty-acid release leads to enhanced hepatic VLDL cholesterol production which correlates with the degree of hepatic fat accumulation. [1, 17] Hyperinsulinemia is also associated with low HDL cholesterol levels. [18, 19]

Ishfaq Ahmed et al [1] in a study showed the high prevalence of dyslipidemia in subjects with type 2 diabetes mellitus and also stated, hypertriglyceridemia is more common than hypercholesterolemia in diabetic subjects. The study by Madhu SV et al [6] demonstrated an altered postprandial response of serum triglycerides following oral fat challenge in male type 2 diabetes subjects compared to controls.

Results from our study have shown that glycosylated hemoglobin and blood glucose level in the diabetic patients are positively co-related. Total cholesterol, serum triglyceride, LDL cholesterol, VLDL cholesterol and total cholesterol/HDL cholesterol ratio were significantly higher in diabetic subjects than healthy controls and HDL cholesterol and HDL cholesterol/LDL cholesterol ratio were significantly lower in type 2 diabetic patients compared to control group, this suggests that type 2 diabetes patients are predispose to the development of cardiovascular complications. Our results are comparable to the studies performed by different scientists in different areas. [20-22] Although one of the studies found the lower level of triglyceride in type 2 diabetic patients. [23]

The Diabetes complications and control trial (DCCT) [24] carried out by National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK), USA, established that, HbA1c is the gold standard of glycemic control. The level of HbA1c value ≤7.0% was said to be appropriate for reducing the risk of cardiovascular complications. [25] It is shown that HbA1c was found to have positive correlation with total cholesterol, LDL cholesterol and triglycerides in diabetic patients. [26]

In the present study, we divided diabetic patients into 2 groups as per the HbA1c cut-off of 7.0%. The diabetic patients with HbA1c value >7.0% exhibited a significant increase in total cholesterol, LDL Cholesterol, Total cholesterol/HDL cholesterol ratio & significantly lower levels of HDL cholesterol/LDL cholesterol ratio, and without any significant alteration in triglycerides & HDL Cholesterol in comparison to patients with HbA1c value ≤7.0%.

Our results regarding total cholesterol and the atherogenic ratios are in accordance with Khan HA et al [27] found no

significant difference in LDL cholesterol, but there was significant alteration found in other lipid parameters in three different groups based on good, poor and bad glycemic control. Improving glycemic control can substantially reduce the risk of cardiovascular events in diabetics. [28] It has been estimated that reducing the HbA1c level by 0.2% could lower the mortality by 10%. [29]

CONCLUSIONS

With higher HbA1c value, severity of dyslipidemia increases in patients. As elevated HbA1c and dyslipidemia are independent risk factors of cardiovascular diseases, diabetic patients with elevated HbA1c and dyslipidemia can be considered as a very high risk group for cardiovascular diseases.

REFERENCES

1. Ishfaq A, Tabassum A, Ganie MA, Syed M: Lipid profile in type II diabetes mellitus patients of Kashmir region. IJEM, 2008, 12(6&7):13-14.
2. Nasir A, Khan J, Siddiqui TS: Frequency of dyslipidaemia in type 2 diabetes mellitus in patients of hazara division. J Ayub Med Coll Abbottabad 2008, 20(2):51-54
3. Uttra KM, Devrajani BR, Syed ZAS, Devrajani T, Das T, Raza S and Naseem: Lipid Profile of Patients with Diabetes mellitus (A Multidisciplinary Study). World Applied Sciences Journal 2011, 12(9):1382-1384
4. Mohammadi H, Malki AE, Hassar M, Bouchrif B, Qarbal F, Dahbi F, Hilal L and Ghalim N: Glycaemic Control, HbA1c, and Lipid Profile in Children with Type 1 Diabetes Mellitus. European Journal of Scientific Research, 2009, 29(2):289-294
5. Mahato RV, Gyawali P, Raut PP, Regmi P, Singh KP, Pandeya DP, Gyawali P: Association between glycaemic control and serum lipid profile in type 2 diabetic patients: Glycosylated haemoglobin as a dual biomarker. Biomedical Research 2011, 22 (3):375-380
6. Madhu SV, Mittal V, Ram BK, Srivastava DK: Postprandial Lipid Abnormalities in Type 2 Diabetes Mellitus. JAPI, 2005, 5:1043-1046
7. Sultana R: Impact of duration of type 2 diabetes mellitus on lipid profile. Gomal Journal of Medical Sciences, 2010, 8(1):57-59
8. Third Report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III) NIH Publication No. (01-3670) May 2001
9. Trinder, P. Glucose assay: A colorimetric enzyme-kinetic method assay. Ann Clin Biochem 1969; 6:24.
10. Jeppsson JO, Kobold U, Barr J, Finke A, Hoelzel W, Hoshino T, Miedema K, Mosca A, Mauri P, Paroni R, Thienpont L, Umemoto M, Weykamp C: Approved IFCC reference method for the measurement of HbA1c in human blood. Clin Chem Lab Med, 2002, 40(1):78-89.
11. Allain CC, Poon LS, Chan CS, Richmond W, Fu PC: Enzymatic determination of total serum cholesterol. Clin Chem, 1974, 20(4):470-475.
12. Warnick GR, Nauck M, Rifai N: Evolution of method for measurement of HDL cholesterol: from ultracentrifugation to homogenous assays. Clin Chem, 2001, 47(9):1579-1596.
13. Bucolo G, David H: Quantitative determination of serum triglycerides by the use of enzymes. Clin Chem, 1973, 19(5):476-482.
14. Friedewald WT, Levy RI, Fredrickson DS: Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. Clin Chem 1972, 18:499-502.
15. Taskinen MR: Diabetic dyslipidaemia: from basic research to clinical practice. Diabetologia, 2003, 46:733–749

16. Krauss RM, Siri PW: Dyslipidemia in type 2 diabetes. Med Clin North Am, 2004, 88:897-909
17. Frayn KN: Adipose tissue and the insulin resistance syndrome. Proc Nutr Soc 2001, 60(3):375-80.
18. Mooradian AD, Albert SG, Haas MJ: Low serum high-density lipoprotein cholesterol in obese subjects with normal serum triglycerides: the role of insulin resistance and inflammatory cytokines. Diabetes Obes Metab, 2007, 9:441–443
19. Mooradian AD, Haas MJ, Wehmeier KR and Wong NC: Obesity-related changes in high density lipoprotein metabolism. Obesity, 2008, 16:1152–1160
20. Garber AJ: Implications of cardiovascular risk in patients wih type 2 diabetes mellitus who have abnormal lipid profile: is lower enough? Diabetes Obese Metab, 2000, 2(5):263-270.
21. Zurro HJ, Hancu N, Lobon CC, Veresiu IA, Suciu L, Remero BE, Dobre L, Jeican D: Pattern of plasma lipids and insulin blood levels in male patient with non-insulin dependent diabetes mellitus and moderate obesity. Rev Clin Esp. 1991, 189(4):172-174
22. Udawat H, Goyal RK, Maheshwari S: Coronary risk and dyslipidemia in type 2 diabetic patients. Assoc Physic India. 2001, 49:970-973.
23. Sharma MD, Pavlik VN: Dyslipidemia in African Americans, Hispanics and whites with type 2 diabetes mellitus and hypertension. Diabetes Obes Metab, 2001, 3(1):41-45.
24. The Diabetes Control and Complications Trial - Implications for Policy and Practice. N Eng J Med, 1993, 329(14):1035-1036.
25. Rohlfing CL, Wiedmeyer HM, Little RR, England JD, Tennill A, Goldstein DE: Defining the relationship be-tween plasma glucose and HbA1c: analysis of glucose profiles and HbA1c in the Diabetes Control and Complications trial. Diabetes Care 2002, 25:275-278.
26. Erciyas F, Taneli F, Arslan B, Uslu Y: Glycemic control, oxidative stress and lipid profile in children with type 1 diabetes mellitus. Arch Med Res 2004, 35:134-40.
27. Khan HA, Sobki SH, Khan SA: Association between glycaemic control and serum lipids profile in type 2 diabetic patients: HbA1c predicts dyslipidaemia. Clin Exp Med 2007, 7:24-29.
28. Selvin E, Wattanakit K, Steffes MW, Coresh J, Sharrett AR: HbA1c and peripheral arterial disease in diabetes: the Atherosclerosis Risk in Communities study. Diabetes Care 2006, 29:877-882.
29. Khaw KT, Wareham N, Luben R, Bingham S, Oakes S, Welch A, Day N: Glycosylated haemoglobin, diabetes, and mortality in men in Norfolk cohort of European Pro-spective Investigation of Cancer and Nutrition (EPIC-Norfolk). Br Med J 2001, 322:15-18.

How to cite this article: Momin AA, Bankar MP, Bhoite GM. Glycosylated hemoglobin (HbA_{1C}): Association with dyslipidemia and predictor of cardiovascular diseases in type 2 diabetes mellitus patients. Int J Health Sci Res. 2013;3(8):40-46.

****