Journal of Cardiovascular Disease Research

ISSN: 0975-3583, 0976-2833 VOL 13, ISSUE 05, 2022

Original Research Article

Relationship Between Serum Homocysteine and Lipid Profile, in Type II Diabetes Mellitus Patients: A Comparative Study

[1]Dr.Dharmesh Gamit, [2]Dr Kaushik Chaudhari, [3]Dr RuhanOza, [4*] Dr.Bhavita Patel, [5]Dr. Punit Patel

[1] Associate Professor, Department of Biochemistry, NAMO Medical Education and Research Institute, Silvassa, Dadra and Nagar Haveli, India

[2] Assistant Professor, Department of Biochemistry, Banas Medical College and Research Institute, Palanpur, Gujarat, India

[3] Assistant Professor, Department of Biochemistry, Pramukh Swami Medical College, Karamsad, Gujarat, India

[4]Assistant Professor, Department of Biochemistry, NAMO Medical Education and Research Institute, Silvassa, Dadra and Nagar Haveli, India

[5]Assistant Professor, Community Medicine Department, Banas Medical College and Research Institute, Palanpur, Gujarat, India.

Corresponding Author: Dr. Bhavita Patel

Abstract

Background: Patients with type II diabetes are a two to four-fold more prone to coronary artery disease than normal healthy persons. Hyperhomocysteinemia have a strong association with the onset of atherosclerosis. Elevated homocysteine in type-II DM causes endothelial cell damage by extravagant sulfation of collagen which increases risk of thrombosis and arteriosclerosis.

Objective:This study was conducted todetermine the relationship between serum homocysteine levels and lipid profile in type 2 diabetes patients.

Materials and Methods: Total 50 newly diagnosed with type II DM patients and 50 apparently healthy controls were enrolled for the present study. Blood samples were collected for estimation of fasting blood sugar, HbA1c, serum homocysteine and lipid profile.

Results: Homocysteine level (Hcy) among diabetic patients was 18.23 ± 3.45 μmol/L which was significantly higher than control group (4.90 ± 1.18 μmol/L, $p < 0.001$). Serum cholesterol, TG, LDL and VLDL were significantly higher among diabetic patients with hyperhomocysteinemia than diabetic patients with normal with S. homocysteine ≤ 15 μmol/L. Positive correlation of homocysteine was observed with and FBS and HbA1c (r- 0.721 and 0.491 respectively)

Conclusion: Type 2 diabetes individuals are at risk of having hyperhomocysteinemia which leads to dyslipidemia. There is a strong positive correlation between serum Hcy and glycemic state, implying that the association between hyperhomocysteinemia and an increased rate of coronary heart disease events among diabetes patients.

Keywords: Coronary heart disease, Diabetes mellitus, Homocysteine, Lipid profile

Introduction

Diabetes Mellitus (DM) is a complex metabolic disease defined by long-term hyperglycemia and caused by a complex interplay of genetic, environmental, cultural, nutritional, and behavioural factors.[1]India is considered the "Diabetic Capital of the World.[2] It is rising

ISSN: 0975-3583, 0976-2833 VOL 13, ISSUE 05, 2022

much more rapidly, presumably because of increasing obesity, reduced activity level as countries become more industrialized, and the aging of the population.[3] Patients with type II diabetes are a two to four-fold more prone to coronary artery disease and post myocardial infraction mortality than normal healthy persons.[4]High Triglyceride levels (TG), low High Density Lipoprotein (HDL) cholesterol, and elevated Low Density Lipoprotein (LDL) cholesterol are characteristics of dyslipidemia associated with type 2 diabetes. Diabetic dyslipidemia, specifically an increase in small dense LDL, increases risk of Coronary Heart Disease (CHD).[5]

Demethylation of methionine produces homocysteine (Hcy) which is a sulfur-containing intermediate amino acid.[6]Hyperhomocysteinemia have a strong association with the onset of atherosclerosis, which manifests as arterial thrombosis and venous thrombosis.[7]Elevated homocysteine in type-II DM increases the production of homocysteine disulfides and homocysteine thiolactone, leading to endothelial cell damage by extravagant sulfation of collagen which increases risk of thrombosis and arteriosclerosis.[8]Homocysteine levels in the blood have been associated with increased lipid peroxidation. The relationship between serum homocysteine levels and type 2 diabetes is still debated. As a result, this study was conducted to determine the relationship between serum homocysteine levels and lipid profile in type 2 diabetes patients, which may be useful in determining the risk of developing CVD and atherosclerosis among diabetic patients.[2]

Materials and Methods

This observational comparative study was undertaken at department Biochemistry and department of General Medicine of tertiary care hospital of Gujarat after receiving approval from institutional ethics committee. Total 50 newly diagnosed with type II DM patients of 18 to 65 years age group were enrolled for the present study. Fifty apparently healthy controls of with matched age and gender were enrolled for the study. Patients with renal, liver disease, congestive heart failure, vascular disease, chronic obstructive pulmonary disease, thyroid dysfunction, neuropathy cancer or on lipid lowering drugs were excluded from the study. A written consent was taken from each participant. A detailed history related to socio demographic profile, addiction and past medical history were taken and physical and systemic examination was performed.

After an overnight fast of 8 hours, 10 ml of venous blood was taken in a) Ethylene diamine tetra acetic (EDTA) vial for glycosylated haemoglobin (HbA1c) measurement. b) Fluoride oxalate vial for estimating fasting blood sugar (FBS). c) Plain vial for homocysteine (Hcy), creatinine, and lipid Profile. The blood samples were centrifuged for 10 minutes at 3000 rpm. The serum was isolated and stored at 2 to 8 degrees Celsius and analysed for homocysteine and lipid profile estimation. Axis-Homocysteine Enzyme Immunoassay (EIA) kit from Axis-Shield Diagnostic, UK was used to determine serum Hcy levels using an ELISA method. Serum lipid profile estimation was done by Enzymatic Colorimetric Test by using Kits Human Diagnostic mbH. Hyperhomocysteinemia was defined as a concentration of Hcy ≥15 µmol/l.[9]

The data was entered in Microsoft excel 2010 and analysed with Epi info 7.1.1. Quantitative data were presented as mean ± SD and compared by applying Student's t-test. P value less than 0.05 was considered significant.

Results

Table 1 shows comparison of socio demographic profile and laboratory parameter between case and control group. Both groups were comparable for age and gender distribution. Smoker were significantly higher in diabetes group (52.0%) as compared to control group

ISSN: 0975-3583, 0976-2833 VOL 13, ISSUE 05, 2022

(24.0%, p – 0.003). Glycemic parameter (FBS, HbA1c) were significantly higher in diabetic patients as compared to control group. Lipid profile parameter such as Cholesterol, TG, HDL, LDL and VLDL were also significantly among diabetic than non-diabetic patients. Serum Homocysteine level (Hcy) among diabetic patients was 18.23 ± 3.45 μmol/L which was significantly higher than control group (4.90 ± 1.18 μmol/L, p < 0.001).

Table 1: Comparison of socio demographic profile and laboratory parameter between case and control group

Parameter	Diabetic Patients (n-50)	Control group (n = 50)	p-value
Age (years)	55.2 ± 13.27	54.9 ± 12.23	0.23
Male: Female	30: 20	30:20	1.00
Smoker	26 (52.0%)	12 (24.0%)	0.003
BMI (kg/m^2)	24.7 ± 4.3	29.1 ± 4.8	< 0.001
FBS (mg/dL)	245.60 ± 69.2	99.14 ± 12.58	< 0.001
HbA1c (%)	11.90 ± 3.45	4.90 ± 1.18	< 0.001
S. Hcy (μmol/L)	18.23 ± 4.33	9.98 ± 2.07	< 0.001
S. Cholesterol (mg/dL)	178.52 ± 34.4	149.14 ± 22.34	< 0.001
S. TG (mg/dL)	149.3 ± 34.5	123.32 ± 22.75	< 0.001
S. HDL (mg/dL)	41.2 ± 6.76	47.24 ± 4.56	0.24
S. LDL (mg/dL)	112.0 ± 22.3	87.44 ±23.4	< 0.001
S. VLDL (mg/dL)	27.23 ± 12.32	25.23 ± 5.56	0.08

The diabetic patients were divided into two groups according to serum Hcy levels. A serum Hcy level of >15.0 μmol/L is considered as hyperhomocysteinemia. Total 38 patients had hyperhomocysteinemia and 12 diabetic patients had homocysteine level <15.0 μmol/L. Higher cholesterol (184.21 ± 35.6 mg/dL) and TG level (155.43 ± 53.01 mg/dL) in diabetic patients with hyperhomocysteinemia than diabetic patients with S. homocysteine ≤ 15 μmol/L (157.23 ± 42.23 mg/dL, 132.24± 76.2 mg/dL respectively). Similarly LDL and VLDL were also significantly higher among diabetic patients with hyperhomocysteinemia than diabetic patients with normal with S. homocysteine ≤ 15 μmol/L.

Table 2: Comparison of lipid profile and gylcemic parameter between normal and hyperhomocysteinemia among patients with Type II DM (n-50)

Variables	S. Hcy> 15 μmol/L (n = 38)	S. Hcy ≤ 15 μmol/L (n = 12)	p-value
FBS (mg/dL)	263.6 ± 52.10	228.81 ± 65.20	0.04
HbA1c (%)	13.32 ± 3.21	9.23 ± 2.34	< 0.001
S. Hcy (μmol/L)	22.23 ± 5.64	13.23 ± 2.13	< 0.001
S. cholesterol(mg/dL)	184.21 ± 35.6	157.23 ± 42.23	0.002
S. TG (mg/dL)	155.43 ± 53.01	132.24± 76.2	0.06
S. HDL (mg/dL)	42.23 ± 5.43	40.21 ± 6.22	0.23
S. LDL (mg/dL)	119.23 ± 24.12	93.33± 33.43	< 0.001
S. VLDL (mg/dL)	32.23 ± 15.23	25.21 ± 10.46	0.04

Positive correlation of homocysteine was observed with and FBS and HbA1c (r- 0.721 and 0.491 respectively)

ISSN: 0975-3583, 0976-2833 VOL 13, ISSUE 05, 2022

Table 3: Correlation of Hcy with FBS and HbA1c

Correlation	r-value	p-value
Hcy and FBS	0.721	< 0.00 1
Hcy and HbA1c	0.491	0.03

Discussion

According to Nickolas et al[10], atherosclerosis in diabetes mellitus is a persistent low-grade inflammatory condition. Hyperhomocysteinemia is a substantial risk factor for atherosclerosis in both diabetic and non-diabetic people. It has the potential to cause chronic vascular complications.[2] Despite numerous studies, the association between hyperhomocysteinemia and cardiovascular risk is still unknown.

In the present study, lipid profile parameters among diabetic group such as cholesterol, TG, LDL, and VLDL, were considerably higher than the control group. Hypertriglyceridemia with elevated LDL particles and low HDL cholesterol levels in diabetic individuals was observed in the study of Sniderman et al.[11] and Choudhary J et al.[2] Insulin resistance and hyperglycemia leads lipid abnormalities. The aetiology of this dyslipidemia could be due to increased secretion of TGs rich in VLDL from the liver and their poor clearance.[12]

In the present study, 76% of the diabetic subjects had serum Hcy>15 μmol/L. Serum Hcy level among diabetic patients (18.23 ± 3.45 μmol/L) was higher than control group (4.90 ± 1.18 μmol/L, $p < 0.001$). Similar finding was observed in the study of Bansal S et al.[13] (Diabetic group : 12.9 ± 5.6 μmol/L, Non diabetic group: 9.9 ± 3.6 μmol/L). Chico et al.[14] found a similar result in their research. According to Puri et al.[15], the mean level of homocysteine in diabetes patients was nearly double that of controls. Homocysteine levels >18 mol/L were found in 72.5% of patients, but in only 26.7 % of controls. On the contrary, Wollesen et al.[16] found that serum Hcy levels in type 2 diabetic patients (10.6 μmol/L) were insignificant ($p>0.05$) when compared to control (11.1 μmol/L).

Serum Hcy levels cause endothelial dysfunction by increasing oxidative stress and inhibits vascular dilatation by reducing the release of nitric oxide (NO). Hyperhomocysteinemia enhances intima-media thickening by stimulating smooth muscle cell proliferation and collagen synthesis. High Hcy levels in the blood are thought to have thrombogenic activity because they modify the coagulation system and encourage platelet aggregation. Increased lipid peroxidation is also linked to elevated blood Hcy levels, which predisposes to atherosclerosis.[13] According to Hoogeveen et al.[17], hyperhomocysteinemia raised the risk of cardiovascular disease by 1.6 times in type 2 diabetes mellitus patients compared to non-diabetic people.

In the present study, significant positive correlation of homocysteine was observed with and FBS and HbA1c (r- 0.721 and 0.491 respectively). Bansal S et al.[13] also observed positive correlation of Hcy with FBS (0.631) and HbA1c (0.416). HbA1c levels were also considerably greater in T2DM patients with hyperhomocysteinemia compared to patients with normal homocysteine levels, according to O A Ala et al.[18] The findings suggest that metabolic regulation may have an impact on Hcy levels.[19]

In the present study, significantly higher values of serum Cholesterol, LDL, and VDL were found among diabetic patients with hyperhomocysteinemia than diabetic patients with normal homocysteine level. Balu et al.[20] found significant difference in serum Hcy levels (15.064 ± 4.82, $p < 0.001$), serum Cholesterol (201.2 ± 29.69, $p < 0.001$), LDL (128.08 ± 29.09, $p < 0.001$), and HDL (40.08 ± 3.29, $p < 0.05$) when compared to controls. The relationship of

ISSN: 0975-3583, 0976-2833 VOL 13, ISSUE 05, 2022

high Hcy with high cholesterol levels observed in this study indicates causal relationship between plasma Hcy and cholesterol. According to Li et al.,[21] high serum Hcy has a positive effect on the hydroxyl methyl glutaryl CoA synthase enzyme and resulting hypercholesterolaemia. Furthermore, they suggested that high Hcy increases cholesterol accumulation in the endothelial cells.

Conclusion

According to the findings of the present study, type 2 diabetes individuals are at risk of having hyperhomocysteinemia which leads to dyslipidemia and vice versa. Homocysteine is a biomarker for endothelial dysfunction and dyslipidemia which are the predictor of cardiovascular complications. There was also a strong positive correlation between serum Hcy and glycemic state, implying that the association between hyperhomocysteinemia and an increased rate of coronary heart disease events among diabetes patients. As a result, serum Hcy is an atherogenic and thrombogenic marker that can be used to detect cardiovascular risk events in T2DM patients at an early stage.

References

1. Ghassibe-Sabbagh M, Deeb M, Salloum AK, Mouzaya F, Haber M, Al-Sarraj Y, Chami Y, Akle Y, Hirbli K, Nemr R, Ahdab R. Multivariate epidemiologic analysis of type 2 diabetes mellitus risks in the Lebanese population. Diabetol Metab Syndr 2014 Dec;6(1)1-2
2. Choudhary J, Fiza B, Sinha M, Mathur R. Serum Homocysteine and Its Association with Lipid Profile in Type II Diabetes Mellitus. Indian J Med Biochem January-June 2018;22(1)26-31
3. Powers AC. Diabetes Mellitus. In: Longo DL, Fauci AS, Kasper DL, Hauser SL, Jameson JL, Loscalzo J, editors. Harrison's Principles of Internal Medicines. 18thed. New YorkMcGraw-Hill; 2008.p2969
4. Deepa R,Arvind K, Mohan V. Diabetes and risk factors for Coronary Artery Disease. Int J Curr Sci 2002;831497- 1505
5. Ramachandran, Snehalata. Epidemiology of heart disease in type 2 diabetes. Curr Sci 2002; 83(12)1
6. Finkelstein JD. Methionine metabolism in mammals. J Nutr Biochem 1990;1228–37
7. Cattaneo M. Hyperhomocysteinemia and thrombosis. Lipids 2001;36:13-26
8. Kannel WB, McGee DL. Diabetes and cardiovascular disease: the Framingham study. JAMA 1979 May 11;241(19)2035-8
9. Platt DE, Hariri E, Salameh P, Merhi M, Sabbah N, Helou M, Mouzaya F, Nemer R, Al-Sarraj Y, El-Shanti H, Abchee AB. Type II diabetes mellitus and hyperhomocysteinemia: a complex interaction. Diabetol Metab Syndr 2017 Dec;9(1)1-7
10. Nikolaos P, Dimitris T, Gerasimos S, et al. Is Fibrinogen a Marker of Inflmmation in Coronary Artery disease? Hell J Cardiol 2010;5341-9
11. Sniderman AD, Scantlebury TH, Cianflone K. Hypertriglyceridemic hyperapo B: the unappreciated atherogenic dyslipoproteinemia in type 2 diabetes mellitus. Ann Intern Med 2001 Sep;135(6)447-59
12. Krauss RM. Lipids and lipoproteins in patients with type 2 diabetes. Diabetes Care 2004 Jun;27(6)1496-1504
13. Bansal S, Kapoor S, Singh GP, Yadav S. Serum homocysteine levels in type 2 diabetes mellitus patients. Int Jr Contemp Med Res 2016;3(11)3393-6
14. Chico A, Perez A, Cardoba A, Arcelus R, Carreras G, Deleiva A, et al. Plasma homocysteine is related to albumin excretion rate in patients with DM: a new link between diabetic nephopathy and cardiovascular disease. Diabetol 1998;41684-93

ISSN: 0975-3583, 0976-2833 VOL 13, ISSUE 05, 2022

15. Puri A, Gupta OK, Dwivedi RN, Bharadwaj RPS, Na-rain VS, Singh S. Homocysteine and lipid levels in young patients with coronary artery disease. J Assoc Phys India 2003 Jul;51681-5
16. Wollesen F, Brattstrom L, Refsum H, Ueland MP, Berglumal L, Berne C. Plasma total homocysteine and cysteine in relation to glomerular filtration rate in diabetes mellitus. Kidney Int 1999;551028-35
17. Hoogenveen EK, Kostense PJ, Jacobs C, Dekker JM, Nijpels G, Heine RJ, et al. Hperhomocysteinemiainceases risk of death, especially in type 2 diabetes: 5-year followup of the Hoorn Study. Circ 2000;1011506-11
18. Ala OA, Akintunde AA, Ikem RT, Kolawole BA, Ala OO, Adedeji TA. Association between insulin resistance and total plasma homocysteine levels in type 2 diabetes mellitus patients in south west Nigeria. Diabetes & Metabolic Syndrome: Clin Res Rev 2017 Dec 1;11S803-9
19. Drzewoski JL, Czupryniak GC, Bald E. Hyperhomocysteinaemia in poorly controlled type 2 diabetes patients. Diabetes Nutr Metab 2000;13312–4
20. Balu MK, Santha K, Santosh kumar N, GnanaDesigan E, Mohammad A. Serum Homocysteine And Lipid Profile Levels in Type 2 Diabetes Mellitus Patients. Int J Med Res Heal Sci 2013;2200-5
21. Li H, Lewis A, Brodsky S, Rieger R, Iden C, Goligorsky MS. Homocysteine induces 3-hydroxy-3-methylglutaryl coenzyme a reductase in vascular endothelial cells: a mechanism for development of atherosclerosis? Circ 2002;1051037–43