Research Article

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Evaluation of Serum Lipid and Cardiac Enzyme Status in Diabetic and Non-Diabetic Adults in Dhaka, Bangladesh.

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ABSTRACT

Hypertension is a leading risk factor for complications in diabetes mellitus patients, and hyperlipidemia is frequently observed among individuals with diabetes. This study compared the lipid profiles and cardiac enzyme levels in diabetic and non-diabetic individuals in Bangladesh, focusing on fasting blood glucose, cardiac enzymes, and lipid profiles. Serum samples were analyzed to measure total cholesterol (TC), HDL-cholesterol (HDL-C), LDL-cholesterol (LDL-C), and triglycerides (TG). Cardiac biomarkers; creatine phosphokinase (CPK), creatine kinase-MB (CK-MB), and lactate dehydrogenase (LDH) were also assessed. A total of 620 diabetic and 520 non-diabetic participants aged 30-60 years were included to examine differences in lipid profiles and cardiac enzyme levels. This study observed mean values of TC ($p=0.003$), TG ($p=0.001$), BMI ($p<0.001$), LDL-C ($p=0.036$), CKMB ($p<0.001$) and LDH ($p<0.001$) were significantly higher in diabetic patients than in the nondiabetic patients. However no significant difference was observed in HDL-C ($p=0.168$) and CPK ($p=0.090$) levels between diabetic and non-diabetic groups.

Keywords : *Diabetes mellitus, Hypertension, Cardiac enzymes, Lipid profile, Hyperlipidemia.*

INTRODUCTION

Diabetes mellitus has become one of the most widespread health concerns in Bangladesh, affecting both urban and rural populations at an increasing rate. It is characterized by elevated blood glucose levels, which can lead to complications in various organs, including the eyes, kidneys, heart, nerves, and blood vessels. Research has shown that oxidative stress plays a significant role in the development of diabetes-related complications, likely through the generation of free radicals [1, 2]. By 2025, the number of individuals with diabetes is projected to double, accompanied by a substantial increase in cardiovascular disease incidence [3]. Cholesterol is an essential component of cell membranes and plasma lipoproteins. It also serves as the precursor for steroid hormone synthesis [4]. Dyslipidemia, or abnormal lipid levels, is commonly seen in individuals with diabetes mellitus and is influenced by insulin resistance and deficiency, affecting enzyme activity and lipid metabolism pathways [5]. Typical lipid abnormalities

in diabetes include elevated triglycerides (TG), decreased high-density lipoprotein cholesterol (HDL-C), and increased low-density lipoprotein cholesterol (LDL-C) [6,7].

The World Health Organization notes that elevated cardiac biomarkers are indicative of myocardial infarction (MI), while epidemiological studies observed abnormalities in lipids and lipoproteins to an increased risk of atherosclerotic cardiovascular disease [8,9]. Other risk factors for coronary heart disease (CHD) include smoking, hypertension, obesity, and diabetes mellitus. This study aimed to compare the lipid profiles and cardiac enzyme levels in diabetic and non-diabetic individuals, examining whether abnormalities in these markers are associated with diabetes.

MATERIALS AND METHODS

Study Design and Participants

This study involved both diabetic and non-diabetic participants from Dhaka, Bangladesh, and was conducted at the Zainul Haque Sikder Women's Medical College Hospital and Cardiac Research Centre, Gulshan Branch, from January to December 2023. Diabetic participants were confirmed cases based on fasting plasma glucose levels and plasma glucose levels 2 hours post-OGTT or post-breakfast, with values exceeding 7.0 mmol/L and 11.0 mmol/L, respectively. Non-diabetic participants were confirmed to be free of diabetes based on overnight fasting plasma glucose levels. After initial interviews, blood samples were collected from all participants to measure fasting blood sugar, lipid profiles (including TC, HDL-C, LDL-C, and TG), and cardiac enzymes such as Creatine Phosphokinase (CPK), CK-MB, and Lactate Dehydrogenase (LDH).

Laboratory Procedures

Personal consent was obtained from all participants, who were asked to fast for 10 to 12 hours before blood collection. Venous blood samples were drawn from both diabetic and non-diabetic individuals. Samples were allowed to clot, and serum was separated by centrifuging at 3,000 rpm for 15 minutes at room temperature. The serum was stored at -20°C until analysis. For lipid profile and cardiac enzyme measurement, 4 ml of blood was collected in a plain test tube and centrifuged, while 1 ml was used for glucose measurement in a tube containing fluoride to preserve the sample.

Lipid Profile Estimation

The lipid profile was analyzed through enzymatic methods. Cholesterol esterase (CE) catalyzes the breakdown of cholesterol esters to free cholesterol, which is further oxidized by cholesterol oxidase, resulting in the formation of cholest-4-ene-3-one and hydrogen peroxide. The concentration of total cholesterol was measured at wavelengths of 540, and 700 nm based on the absorbance of oxidized N.N-diethylaniline-HCl/4-aminoantipyrine (DEA-HCl/AAP) [10].Triglyceride levels were measured through an enzymatic method that employs various enzymes to determine the glycerol content in the sample. The absorbance change due to quinoneimine formation correlates with the glycerol amount and was recorded at 510 and 700 nm [11,12].HDL-C was measured by isolating other lipoproteins (chylomicrons, VLDL, LDL) in a water-soluble complex formed with dextran sulfate in the presence of magnesium sulfate. HDL cholesterol is oxidized, producing 4-cholestenone and hydrogen peroxide, which then reacts to produce a colored dye measured bichromatically at 600/700 nm [13,14].

LDL-C levels were calculated using a bichromatic endpoint technique (540, 700 nm) based on color intensity, which directly correlates with LDL-C concentration. For samples with triglyceride levels below 400 mg/dl, the Friedewald formula (LDL-C = TC – (TG/5 + HDL-C)) was used [15,16].

Cardiac Enzyme Analysis

Creatine kinase (CK) activity, measured in patient samples, catalyzes the reversible transphosphorylation from phosphocreatine to ADP, producing ATP. The rate of NADPH formation correlates with CK activity and was measured at 340 and 540 nm [17,18].The lactate dehydrogenase (LDH) method used L-lactate as a substrate buffered at pH 9.4. LDH catalyzes oxidation to pyruvate and NADH, absorbing light at 340 nm. LDH concentration was determined as a rate reaction at 340/700 nm, directly proportional to LDH activity in the sample [19,20]. CK-MB, one of the isoform of CPK, measurement is crucial for diagnosing myocardial infarction [21].

Height and Weight Measurement

Participants' weight was recorded to the nearest kilogram using a standardized weighing scale, calibrated regularly. Height was measured with participants standing barefoot, feet together, using a height scale with a steel tape and a horizontal bar that was gently placed on the head to record height accurately [22,23].

Body Mass Index (BMI)

BMI was calculated as weight in kilograms divided by the square of height in meters (kg/m^2). Patients were considered obese if their BMI exceeded 27.8 for males and 27.3 for females [24].

Statistical Analysis

Data analysis was performed using SPSS version 17 (Chicago, IL, USA). A p-value of <0.05 was considered statistically significant. Pearson correlation coefficients were calculated to analyze the relationship between fasting blood sugar and

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other diabetic and non-diabetic variables. Multiple linear regression and partial correlation analyses were conducted, adjusting for covariates, and results were presented as mean ± standard deviation (SD).

RESULTS

This study included a total of 1,140 participants, with 620 diabetic (54.38%) and 520 non-diabetic (45.61%) individuals. The average age for non-diabetic participants was 44.65 ± 8.57 years, while for diabetic participants it was 45.58 ± 7.84 years, with ages ranging from 30 to 60 years in both groups. Body mass index (BMI) was also analyzed, showing an average of 23.39 ± 1.48 kg/m² for non-diabetic subjects and 24.45 ± 2.60 kg/m² for diabetic subjects.Lipid profiles (including TC, HDL-C, LDL-C, and TG), cardiac enzymes (CPK, CK-MB, and LDH), and fasting blood sugar levels were measured in both diabetic and non-diabetic participants. As shown in Table 1, the mean (±SD) values for fasting blood sugar, TC, LDL-C, TG, CK-MB, and LDH were significantly higher in diabetic participants compared to non-diabetic ones. However, HDL-C (p=0.168) and CPK (p=0.09) did not show statistically significant differences between the two groups. The mean serum fasting blood sugar (p<0.001) was notably higher in diabetic individuals (9.84 ± 4.48 mmol/L) compared to non-diabetic individuals (5.54 ± 0.58 mmol/L).

Table 1. Mean values of descriptive characteristics and Lipid & cardiac enzymes of the non diabetic and diabetic subjects.

Name of Biochemical parameters	Non-Diabetic Mean ± SD	Diabetic Mean ± SD	p-value
No. of subjects	620	520	
Age (Years)	44.65 ± 8.57	45.58 ± 7.84	0.083
BMI(kg/m²)	23.39±1.48	24.45±2.60	<0.001
Fasting blood sugar (mmol/L)	5.54 ± 0.58	9.84 ± 4.48	<0.001
Total Cholesterol (mg/dl)	178.68 ± 33.99	193.51 ± 41.40	0.003
HDL-Cholesterol (mg/dl)	39.51 ± 4.16	39.61 ± 4.81	0.168
LDL-Cholesterol (mg/dl)	106.48 ± 29.65	117.57 ± 35.28	0.036
Triglyceride (mg/dl)	161.66 ± 71.46	191.92 ± 93.91	0.001
CPK (U/L)	141.43 ± 36.91	173.58 ± 58.37	0.090
CK-MB (U/L)	15.37 ± 4.65	19.22 ± 6.75	<0.001
LDH (U/L)	339.99 ± 61.87	388.77 ± 82.58	<0.001

Data presented are mean ± standard Deviation (SD). P-value obtained from Independent-Samples "t" test.
In the univariate analysis, HDL-C and LDL-C were not significantly associated with fasting blood sugar levels, whereas TC, TG, CPK, CK-MB, and LDH had significant odds ratios (Table 2). After adjusting for age, sex, and BMI, the odds ratios for TC, TG, CPK, CK-MB, and LDH were 0.936 (0.145 to 0.469), 1.332 (0.165 to 1.486), 3.381 (0.168 to 2.267), 5.138 (0.253 to 0.399), and 3.937 (0.195 to 3.907) respectively, maintaining a significant association with fasting blood sugar levels. When adjusted for these covariates, the associations between TC, TG, CPK, CK-MB, and LDH and fasting blood sugar remained statistically significant (p=0.048, p=0.003, p=0.01, p=0.001, and p=0.001, respectively).

Table 2. An association between fasting blood sugar with biochemical parameters lipid profile and cardiac enzymes effects on non-diabetic and diabetic patients.

Dependent variable	Independent variable	
	FBS	
	Unadjusted	Adjusted
Total cholesterol β-Coefficient (95% CI) p-value	0.939 (0.148 to 0.473) 0.042	0.936 (0.145 to 0.469) 0.048
HDL- cholesterol β-Coefficient (95% CI) p-value	0.245 (0.013 to 0.014) 0.807	0.242 (0.012 to 0.013) 0.809
LDL- cholesterol β-Coefficient (95% CI) p-value	0.924 (0.048 to 0.400) 0.356	0.920 (0.046 to 0.397) 0.367

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Triglyceride β-Coefficient (95% CI) p-value	 1.338 (0.169 to 1.491) 0.002	 1.332 (0.165 to 1.486) 0.003
CPK β-Coefficient (95% CI) p-value	 3.387 (0.172 to 2.272) 0.001	 3.381 (0.168 to 2.267) 0.01
CK-MB β-Coefficient (95% CI) p-value	 5.149 (0.257 to 0.402) < 0.001	 5.138 (0.253 to 0.399) 0.001
LDH β-Coefficient (95% CI) p-value	 3.942 (0.199 to 3.913) < 0.001	 3.937 (0.195 to 3.907) 0.001

P-values were from Multivariate linear regression, adjusted for age, sex and BMI. β- Standard regression coefficient.

Correlation analysis showed that fasting blood sugar was significantly correlated with TC, LDL, CPK, CK-MB, LDH, and BMI. Additionally, TC was significantly correlated with HDL, LDL, TG, CPK, CK-MB, and LDH, while HDL showed a significant correlation with LDL, TG, and CPK. LDL was significantly correlated with LDH, and CPK showed a notable association with CK-MB and LDH. Other significant correlations included CK-MB with LDH and LDH with BMI (Table 3).

Table 3. Pearson's Correlation between different lipid, cardiac enzymes and fasting blood sugar among non-diabetic (520) and diabetic (620) subjects.

		TC	**HDL**	**LDL**	**TG**	**CPK**	**CK-MB**	**LDH**	**BMI**
FBS	r	0.119	0.023	0.099	0.104	0.154	0.248	0.161	0.087
	p-value	0.001	NS	0.005	0.003	<0.001	<0.001	<0.001	0.013
TC	r		0.314	0.648	0.277	0.102	0.107	0.133	0.010
	p-value		<0.001	<0.001	<0.001	<0.001	0.003	<0.001	NS
HDL	r			0.317	0.093	0.101	0.030	0.058	0.015
	p-value			<0.001	<0.001	0.005	NS	NS	NS
LDL	r				0.057	0.048	0.055	0.092	0.044
	p-value				NS	NS	NS	0.009	NS
TG	r					0.091	0.154	0.113	-0.002
	p-value					0.009	<0.001	0.001	NS
CPK	r						0.220	0.197	0.085
	p-value						<0.001	<0.001	0.014
CK-MB	r							0.264	0.015
	p-value							<0.001	NS
LDH	r								0.060
	p-value								0.047

NS = Not significant; FBS = fasting blood sugar; TC = total cholesterol; LDL = low-density lipoprotein; HDL = high-density lipoprotein; TG = triglycerides; BMI = body mass index; CPK = creatinine phosphokinase; LDH = lactate dehydrogenase.

DISCUSSION

This study evaluated and compared the biochemical markers, including commonly used cardiac biomarkers, between individuals with and without diabetes. As a cross-sectional study, it primarily highlights associations between blood lipid levels and cardiac enzymes rather than causal relationships. Consistent with previous studies, we observed significantly elevated levels of the cardiac enzymes CPK, CK-MB, and LDH in diabetic patients. This increase is generally attributed to myocardial tissue injury, where enzymes leak from the cytosol following an infarction. We also observed significant elevations in total cholesterol, LDL cholesterol, and triglycerides in diabetic individuals. LDL cholesterol, which is responsible for transporting cholesterol from the liver to peripheral tissues, is particularly notable. Prior research suggests that high LDL levels increase the risk of atherosclerosis, as LDL is prone to oxidation under certain conditions, leading to the formation of atherosclerotic plaques in the coronary arteries [25]. Brown and colleagues (1996) identified high LDL cholesterol as a key indicator of atherosclerotic

risk [26], supporting the findings of this study, which also associates high LDL cholesterol levels with diabetes. Thus, it is evident that LDL cholesterol acts as an independent risk factor for coronary heart disease (CHD). Elevated blood glucose was also observed in diabetic subjects, a contrast to non-diabetic individuals who showed no significant increase. It is well-documented that diabetes significantly raises the risk of cardiovascular diseases. While diabetes is associated with microvascular complications such as nephropathy and retinopathy, its macrovascular complicationsincluding coronary artery disease, cerebrovascular disease, and peripheral vascular diseaseare among the primary causes of mortality in diabetic populations [27]. The Diabetes Control and Complications Trial (1993) demonstrated that intensive blood glucose management can reduce complications, though not entirely prevent them, indicating the potential benefit of alternative treatments. Research suggests that hyperglycemia-induced oxidative stress, largely due to free radical production, plays a key role in the progression of diabetes and its complications. This has led to interest in antioxidant therapies as a means of mitigating these complications [28]. In this study of Bangladeshi adults, fasting blood glucose showed positive correlations with lipid levels and cardiac enzymes. As one of the first studies of its kind in this population, further longitudinal research with larger sample sizes is recommended to confirm these associations. However, several limitations are acknowledged like socio-demographic and clinical variables were not thoroughly included, which may have restricted the depth of analysis. Furthermore, a more comprehensive approach could have considered additional factors such as patients' confirmed diagnoses, nutritional and habitual status, Troponin-I levels, and family medical history, all of which could be vital in exploring the relationships between cardiac enzymes and diabetic conditions. Ultimately, the findings suggest that diabetes poses an increased risk for coronary heart disease and hyperlipidemia.

CONCLUSION

This study found that elevated fasting blood glucose levels positively correlate with serum levels of cardiac enzymes and lipid profiles. These findings highlight the importance of routine lipid profiling for individuals with diabetes mellitus (DM). Monitoring glycosylated hemoglobin could further aid in assessing diabetes control. Primary preventive measures, including the adoption of healthy lifestyle habits, should be emphasized for all diabetic individuals to reduce the risk of coronary heart disease (CHD) and atherosclerosis. Future research is needed to examine dietary patterns among diabetic patients in Bangladesh, as well as other potential components to hyperlipidemia. Integrating the analysis of cardiac enzymes and lipid profiles may offer a valuable approach to managing coronary heart disease, hyperlipidemia, and atherosclerosis in patients with diabetes.

REFERENCES

1. Nogichi, H., 2007. Stem cells for the treatment of diabetes. Endocr. J. 54, 7–16.
2. Manna, P., Sinha, M., Sil, P.C., 2009. Protective role of arjunolic acid in response to streptozotocin-induced type-I diabetes via the mitochondrial dependent and independent pathways. Toxicology 257, 53–63.
3. King H, Aubert RE, Herman WH., 1998. Global burden of diabetes, 1995-2025: prevalence numerical estimates, and projections. Diabetes Care 21:1414-31.
4. Ganong, W., 1979. Energy balance, metabolism and Nutrition- Rev. Med. Physiol. (9th edition), pp 239 -240.
5. Gibbons GF., 1988. Hyperlipidaemia of diabetes. Clin Sci. 71:477–86.
6. Beckman JA, Creager MA, Libby P., 2002. Diabetes and atherosclerosis, Epid, Pathophysiology, and management: Review article. JAMA 287(19):2570–81.
7. Navab M, Berliner JA, Watson AD, Navab M, Berliner JA, Watson AD, et al., 1996. The yin and yang of oxidation in the development of the fatty streaks: a review based on the 1994 George Lyman Duff Memorial Lecture. ArteriosclerThrombVasc Biol. 16:831–42.
8. Lansky AJ, Stone GW., 2010. Periprocedural myocardial infarction: prevalence, prognosis, and prevention. Circ Cardiovasc Interv3(6):602-10.
9. Kashyap ML., 1997. Cholesterol and atherosclerosis: a contemporary perspective. Ann Acad Med Singapore 26(4):517-23.
10. E. Heinz., 1996. Plant glycolipids: structure, isolation and analysis. In advances in lipid methodology Christie Oily Press,3, 211-332.
11. R.A. Coleman, D.P. Lee., 2004. Enzymes of triacylglycerol synthesis and their regulation. Prog. Lipid. Res., 43, 134.176.
12. P.T. Ivanova, S.B. Milne, M.O. Byrne, Y. Xiang, H. A. Brown., 2007. Glycerophospholipid identification

and quantitation by electrospray ionization mass spectrometry. Meth.Enzymol, 432, 21.57.

13. E. Fahy, S. Subramaniam, H.A. Brown., 2005. A comprehensive classification system for lipids. J. Lipid Res.,46(5), 839.61.

14. The Nomenclature of lipids., 1976. Recommendations. European Journal of Biochemistry,79(1), 11.21.

15. Roger Johnsona, Prudence McNutt, Stephen MacMahon1, Richard Robson2., 1997. Use of the friedewald formula to estimate LDL-cholesterol in patients with chronic renal failure on dialysis. Clinical Chemistry,43, 2183-2184.

16. Freidewald, W.T., Levy, R.I., Fredickson, D.S., 1972. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. Clin. Chem. 18, 499–502.

17. G.Schumann., 2002. Clin.Chem.Lab Med., 40, 635-642.

18. G.Schumann., 2003. Clin.Chem.Acta, 327, 69-79.

19. Z.Klin., 1970. Chem.Klin.Biochem,8, 658.

20. AndriesJ.Bakker, BidjaiperkashMirchi, Johannes T. Dijkstra, FreekReitsma, Haye Syperda, AppieZijlstra., 2003. IFCC method for lactate dehydrogenase measurement in heparin plasma is unreliable. Clinical Chemistry, 49, 662-664.

21. I.M.Morisok, J.es.Clayson., 1988. Fine J.S.Clin.Chem,34, 535.

22. Verma BL, Kumar A, Srivastava RN., 1982. Measurement of body build based on weight/height, an index for adults in an Indian population. Ind J Pub Health 26.

23. Frisancho AR., 1984. New standards of weight and body composition by frame size and height for assessment of nutritional status in adults and elderly. Amm J Clin Nutr 40: 808.

24. Olefsky JM. Diabetes mellitus, In cecil textbook of Medicine 18th Ed. Wynagaarden and Smith Jr. Eds; WB Saunders Int ED. 1992; 1291

25. Steinberg D., 1997. Low density lipoprotein oxidation and its pathobiological significance. Biol Chem 272:20963–68.

26. Brown BG, Fuster V., 1996. The impact of management in the stabilization of coronary disease. Lippincott-Raven 191–205.

27. Laakso, M., 1999. Hyperglycemia and cardiovascular disease in type 2 diabetes. Diabetes 48 (5), 937–942.

28. Heibashy, M.I.A., Abdel-Moniem, A.E., 2005. Potential benefits of some antioxidant nutrients in reducing the high levels of some biochemical variables associated with induced hypertension in rats. Isotope Rad. Res. 37 (2), 465–479.