Role of Serum Lipids and Free Radicals in Myocardial Infarction in NIDDM: A Case Control Study

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Abstract**—***Abnormalities that characterizes lipoprotein metabolism in non-insulin dependent diabetes mellitus (NIDDM) patients, fasting concentration of triglyceride rich lipoprotein especially very low density lipoprotein (VLDL) are higher and those of HDL, commonly measured as HDL-c, are lower than among people without diabetes, which leads to increased triglyceride HDL-c ratio and insulin resistance. This type of diabetic dyslipidemia is a major cause of oxidative stress which promote and accelerate atherosclerosis and thus, end organ damage AMI. This present study was carried at the Central Clinical Laboratory MIMSR Medical College Latur, with the aim to find out the role of lipoprotein-triglyceride in myocardial infarction in NIDDM. For this study, patient with myocardial infarction with NIDDM were selected after admitting in MIMSR Medical College Latur. These 25 cases were included in study group and age-matched to these cases 50 healthy subjects were selected as Control group. The lipid profile and total serum lipid peroxides (malondialdehyde) of study and control groups were assessed & compared. It was found that in the control group mean values of total cholesterol was 180.21 ± 18.13 mg % whereas it was 229.21± 23.58 in study group, which was significantly higher in study group. Likewise, mean Serum Triglycerides and Serum Lipid Peroxides (MDA) of study group were also found significantly ($p<0.001$) higher that of control group (228.14 v/s 99.9 and 410.22 v/s 180.96 respectively). It was also revealed in this study that mean Serum HDL-Cholesterol was found significantly lower in study group whereas LDL-Cholesterol (28.72 v/s 53.83) and VLDL-Cholesterol were found significantly higher in study group that control group (150.61 v/s 106.60 and 46.30 v/s 19.8). So it can be concluded that AMI patients with NIDDM have higher Total Serum Cholesterol, Serum Triglycerides, Serum Lipid Peroxides (MDA), LDL- Cholesterol and VLDL-Cholesterol with lower HDL- Cholesterol.*

Keywords: Diabetic dyslipidaemia, Insulin Resistance, Oxidative Stress Malanodialdehyde and Myocardial Infarction.

I. INTRODUCTION

The major independent risk factors for the development of atherosclerosis are the plasma cholesterol concentration, cigarette smoking, hypertension and diabetes, which are by them self's risk factor for coronary heart disease[1]. Diabetes mellitus is a major risk factor for coronary artery disease and is associated with a higher incidence of acute myocardial infarction (AMI) and sudden death. Morbidity, mortality and re-infarction rate are higher following AMI in diabetic than non diabetic patients. Despite recent decline in cardiovascular mortality, atherosclerotic disease is still a major health problem. Incidence of coronary heart disease has shown upward trends in Indian in last decade.[2, 3] A large amount of epidemiological evidences also supports the relationship between serum low density lipoprotein cholesterol (LDL-c) and coronary artery disease (CAD) in Indians.[4] Serum high density lipoprotein-cholesterol (HDL-c) level has been found to have inverse relationship with the CAD.[5] Diabetes mellitus is a common among Indians with CHD both in their land of origin and abroad.[6] Individuals with NIDDM are more likely to have multiple risk factors for CHD than age matched non diabetic subjects. Peoples with diabetes have a risk of CHD two to five times that of non-diabetic individuals.[7,8,9]

Abnormalities that characterizes lipoprotein metabolism in non insulin dependent diabetes mellitus (NIDDM) patients, fasting concentration of triglyceride rich lipoprotein especially very low density lipoprotein (VLDL) are higher, and those of HDL, commonly measured as HDL-c, are lower than among people without diabetes.[10,11] NIDDM is an integral component of the metabolic syndrome.[12] In addition to triglycerides levels, overweight and plasma triglyceride to HDL-c ratio of three or greater as a reliable indicator of insulin resistance.[13] In fact Mc Laughlin & Colleagues[13] suggested that an elevated TG to HDL-c ratio may be "clinically appealing marker because of its robust association with cardio vascular disease (CVD).[14] NIDDM is associated with increase in plasma triglyceride and decrease in plasma HDL-c concentration i.e. dyslipidaemia changes that have been identified as the increasing risk, of AMI. Hyperglycemia, a hallmark of diabetic condition depletes natural antioxidants and facilitates the production of reactive oxygen species (ROS) which has the ability to react with all biological molecules like lipids, proteins, carbohydrates, DNA etc and exert cytotoxic effects on cellular components.[15] Thus, increased ROS and impaired antioxidant defense contributes for initiation and progression of micro and macro vascular complications in diabetis.[16] As ROS and oxygen derived free radicals interact with lipid bilayer of cell membrane, and lipoproteins resulting in the lipid peroxidation.[17] Malanodialdehyde is stable product of lipid peroxidation.[18] Because of this lipoproteins get oxidzed and become atherogenic. A plethora of adverse physiological consequences of elevated MDA levels includes leakiness of cell membranes by altering structural integrity of membrane; inactivation of membrane bound enzymes, inactivation of surface receptor molecules leading to cell-regulating errors, the involvement of oxidized LDL in the foam cell formation leading to atherosclerosis has been documented. Although elevated levels of serum MDA have been reported in type 2 diabetes, there is paucity of literature on the extent of serum MDA levels in type 2 diabetes cases with myocardial infarction, a macrovascular complication. Since the classic risk factors do not account for the excess risk of atherosclerosis in NIDDM, we need new approaches to explain the connection of e risk factors and accelerated vascular disease. Such studies may provide greater insight in the role of oxidative stress along with other predisposing factors in the preventation and precipitation of events like myocardial infarction and the need to use anti-oxidants, omega-3 and life style modifaction as a prophylactic step. Thus, this present study was conducted with the aim to find out the role of lipoprotein-triglyceride in myocardial infarction in NIDDM.

II. METHODOLOGY

A case-control analytic type of observational study was carried out on 25 diabetic patients with myocardial infarction and age & body mass index matched 50 healthy subjects.

Patient belonging to study group were selected among admitted patients in hospital MIMSR Medical College, Latur and diagnosed as diabetic with myocardial infarction. All patients belonging to group II had diabetes with myocardial infarction. Criteria of diagnosis of diabetic were: as all of them were taking insulin or oral hypoglycemic agents or fasting blood sugar levels above 140mg% and who had suffered an attack of myocardial infarction confirmed by elevated CPK-MB and ECG changes.

Subjects for control group were selected from medical, paramedical staff and general public who were around 40 to 60 year of age. All subjects were belonged to the Latur district of Marathwada region. These healthy subjects were nonsmokers, nonobese, nonalcoholic and free from any disease and not taking any drugs that alter lipid and carbohydrates metabolism.

All subjects after taking informed consent were interrogated and detailed examination was done. Blood samples drawn after an overnight fast immediately after admission of diabetic with AMI patient in hospital and at earliest in case of healthy subjects. After serum separation the analysis was done on the same day. Serum triglycerides were estimated by enzymatic method (Autopack Siemens kit) and total cholesterol by enzymatic methods (Autopack Siemens kit) HDL-c measured by phosphotung state method (Autopack Siemens kit). LDL-c and VLDL- c values were calculated by Friedwald's equation[19]. Total serum lipid peroxides were also estimated by reaction with thiobarbituric acid using the method of Nadiger and Chandrakala[20]. Reaction involved in this method is malanodialdehyde combines with thiobarbituric acid to form malanodialdehydethiobarbituric acid complex which is measured colorimetrically at 530nm. The total serum lipid peroxides were calculated by using the coefficient of malanodialdehyde 1.5×10^5 and was expressed as malanodialdehyde /dl serum.

III. RESULTS

In this present study, mean Total Serum Cholesterol of study group was found 229.21 with SD 23.58 whereas of control group 180.21± 18.13. This variation was found with significant difference($p<0.001$). So it can be concluded that study group had significantly higher Serum cholesterol than control group. (Table 1)

Likewise, mean Serum Triglycerides of study group was found significantly ($p<0.001$) higher that of control group (99.9 v/s 228.14). (Table 1)

Likewise, mean Serum Lipid Peroxides (MDA) of study group was found significantly ($p<0.001$) higher that of control group (180.96 v/s 410.22). (Table 1)

Table 1
Comparison of Biochemical parameter in Study and Control group

S. No.	Variables	Control Group (N=50) Mean ± SD	Study Group (N=25) Mean ± SD	Unpaired 't' Test P Value LS
1	**Total Cholesterol (mg%)**	180.21± 18.13	229.21±23.58	-9.959 at 73 DF <0.001 **S**
2	**Triglycerides (mg%)**	99.90±16.14	228.14±52.68	-15.878 at 73 DF <0.001 **S**
3	**Lipid Peroxides (MDA) (nM/dl)**	180.96±35.16	410.22±108.56	-13.648 at 73 DF <0.001 **S**

It was also observed in this present study that among various types of Serum Cholesterol, HDL-Cholesterol of study group was found significantly ($p<0.001$) lower that of control group whereas LDL-Cholesterol and VLDL- Cholesterol of study group was found significantly ($p<0.001$) higher that of control group. (Table 2).

Table 2
Comparison of various types of Cholesterol in Study and Control group

S. No.	Variables	Control Group (N=50) Mean ± SD	Study Group (N=25) Mean ± SD	Unpaired 't' Test P Value LS
1	**HDL-Cholesterol (mg%)**	53.83±16.42	28.72±6.53	7.341 at 73 DF <0.001 **S**
2	**LDL- Cholesterol (mg%)**	106.60±18.2	150.61±22.45	-9.121 at 73 DF <0.001 **S**
3	**VLDL- Cholesterol (mg%)**	19.8±4.6	46.30±11.8	-13.969 at 73 DF <0.001 **S**

IV. DISCUSSION

The catabolism of triglyceride-rich lipoproteins is initiated by lipoprotein lipase, an endothelial enzyme that hydrolyses the triglyceride moiety of chylomicrons and VLDL, and releases fatty acids for energy production in muscle and for storage in adipose tissue. The activity of this enzyme is generally lower in NIDDM patient than in non diabetic people of similar age and degree of adiposity: The difference is more striking for patient with both NIDDM and coronary artery disease (CAD). Lipoprotein lipase activity is low in untreated or poorly controlled NIDDM and increase with improved glycaemic control. In NIDDM passage of triglyceride- rich lipoproteins through the lipolytic cascade is delayed for two reasons: there is a shortage of catalytic sites on lipoprotein lipase and over production of triglyceride saturates the sites that are available. Both mechanisms promote hypertriglyceridaemia. The two components of diabetic dyslipidaemia, high concentrations of triglyceride-rich lipoproteins and low concentrations of HDL, are closely interwoven. Hypertriglyceridaemia contributes to low HDL concentrations in one or combination of following reasons. 1) The first process involves the transfer of surface remnants'-redundant phospholipids and apolipoproteins from lipolysis of triglyceride – rich lipoproteins – to HDL particles. Because lipoprotein lipase activity is decreased and lipolysis impaired in NIDDM, there are fewer surface remnants available to be incorporated into the HDL particle. 2) The large amount of triglyceride-rich lipoproteins and their prolonged residence time in the circulation increased the exchange (mediated by cholesteryl-ester transfer protein) of esterified cholesterol from HDL to triglyceride-rich lipoproteins and of triglyceride to HDL particles. The result is enrichment of the HDL particle core with triglyceride. Enriched HDL has a faster catabolic rate than normal HDL which leads to a lower number of circulating HDL particles. Furthermore, the HDL particles in NIDDM are smaller owing to a high hepatic lipase activity-another feature of NIDDM. Lowered plasma HDL-c and elevated plasma triglyceride levels are features of NIDDM dyslipidaemia (TG/HDL-c ratio above 3) which is a reliable predictor of insulin resistance. Insulin resistance a syndrome that favors atherosclerosis and thus, myocardial infraction. Relying on LDL-c or total cholesterol alone can be misleading. It is also proved that people with obesity, metabolic syndrome or diabetic lipid disorders often have raised triglycerides, low HDL-c and normal or closed to normal LDL-c. Free radicals controls oxygen transport, require for cytochrome P-450 activity, prostaglandin cascade, phagocytosis, blood pressure regulation and detoxication processes. Under certain normal conditions oxygen may accept only one electron (usually in the electron transport chain accepts four electron and get converted to water) and this results in the formation of oxygen derived free radical superoxide. (O2-) which may initiate the chain reaction of free radical formation and lipid peroxidation. Malanodialdehyde is stable product of lipid peroxidation. Elevated malanodialdehyde levels are indicative of lipid peroxidation (oxidative stress). Malanodialdehyde is regarded as a marker of inflammation induced by free radical injury on membrane lipids. Proposed pathway for free radical formation and development of complications due to free radical stress. Diabetes contributes to atherosclerosis through various mechanism such as accelerated formation of reactive oxygen species due to decreased in activity superoxide dismutase, glutathione peroxidase,[21] oxidation of lipoproteins, decreased levels of HDL and autooxidation of glucose because of hyperglycemia. Some studies have shown that diabetes mellitus may result from oxidative injury to the islets due to free radical production catalyzed by decompartmentalized transition metals such as iron and copper.[22] Elevated levels of free radicals in NIDDM oxidizes the lipoproteins. Oxidized lipoproteins particularly oxdized LDL is more atherogenic. Several studies have suggested that the LDL may oxidized in arterial wall and thus, initiate and promote

atherosclerosis.[23] A short lag phase for the oxidation of LDL is associated with coronary atherosclerosis in patients with CHD. Susceptibility of LDL to oxidation has been related to progression of atherosclerosis in carotid and femoral arteries and a higher proportion of partially oxidized LDL was found in patients with progression of atherosclerotic plaque. In addition to this, insulin at physiological level has antiatherogenic effects in vasculature. Many reports show that in NIDDM activity of insulin to induce vasodilation is low due to insulin resistance. Atherosclerosis in NIDDM may be due to multiple factors such as diabetic dyslipidaemia, metabolic syndrome insulin resistance, hypertension, oxidative stress which may act together or independentialy and promotes atherosclerosis and end organ damage myocardial infarction. In our study triglycerides VLDL-c, LDL-c and MDA positively related i.e significantely elevated as compared to healthy subjects. M. Deepa et. al and Rashida Meharan et. al reported that the diabetic dyslipidaemia and elevated free radicals (MDA) have role in advancement of atherosclerosis to myocardial infarction.[24, 25] Observations of this present study are also correlated with these studies.

V. CONCLUSION

It can be concluded that AMI patients with NIDDM have higher Total Serum Cholesterol, Serum Triglycerides, Serum Lipid Peroxides (MDA), LDL- Cholesterol and VLDL- Cholesterol with lower HDL- Cholesterol. For realization of exact role of lipid profile further higher level studies should be done.

CONFLICT OF INTEREST

None declared till now.

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