Comparison of Serum Lipoproteins with Lipidogram and Lp(a) levels in Type 2 Diabetes Mellitus

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Abstract: **This study compares the serum lipoproteins with Lipidigram and Lp(a) levels in Type 2 Diabetes mellitus. More precise identification of serum lipoprotein components is needed for presumptive diagnosis of Type III hyperlipoproteinemia disorder and management of the patients. Serum lipoprotein electrophoresis is a reliable and accurate method which can be used in clinical laboratory for screening abnormal serum lipoproteins in Type 2 DM patients and when a more presumptive diagnosis of phenotypes is needed, it could be an investigation of choice.**

Keyword: Lipoprotein electrophoresis, Lipidogram, Lp(a), Type 2 Diabetes Mellitus, Hyperlipidemia.

INTRODUCTION

Diabetes Mellitus is the most common endocrinological disorder characterized by chronic hyperglycaemia[1,2,17]. Type 2 DM is associated with a cluster of interrelated serum lipid and lipoproteins abnormalities. Core components of diabetic dyslipidemia are increased plasma triglycerides, low concentrations of HDL and predominance of LDL[3,4,5]. Serum Lipoproteins are studied by electrophoresis technique. According to the Electrophoretic mobility, serum lipoproteins are classified into four main fractions: Chylomicrons, Alpha fraction (HDL), Pre-Beta fraction(VLDL) and Beta fraction(LDL)[6].

MATERIAL AND METHODS

90 subjects included in the present study, out of which 60 patients of Type 2 Diabetes Mellitus in age group of 40-65 years of either sex, on oral hypoglycaemic drugs, and 30 normal healthy individuals, age & sex matched from the same population, with no history of diabetes. These 90 subjects were divided into 3 groups:

GROUP A- Normal healthy individuals both males and females in age group of 40-65 years from the general population who volunteered for getting included in the present study

GROUP B- Patients of Type 2 DM (Non Insulin Dependent Diabetes Mellitus) both males & females in the age group of 40-65 years on oral hypoglycaemic drugs with HbA1c <7%

GROUP C - Patients of Type 2 DM (Non Insulin Dependent Diabetes Mellitus) both males & females in the age group of 40-65 years on oral hypoglycaemic drugs with HbA1c >7%

Serum levels of fasting blood sugar, HbA1c, lipoprotein electrophoresis, serum Lp(a) and lipid profile were estimated in all the subjects under case control prospective study.

Fredrickson's classification of hyperlipidemia is used to group disorders of lipoproteins in this study.

Fredrickson's classification of Hyperlipidemia[7]

Type	Lipoprotein Elevated	Cholestrol	Triglyceride	Risk of Atherosclerosis
I	Chylomicrons	+	+++	Not elevated
IIa	LDL	++	Normal	High
IIb	LDL + VLDL	++	++	High
III	IDL	++	++	Moderate
IV	VLDL	+	++	Moderate
V	Chylomicrons + VLDL	+	++	Not elevated

EXCLUSION CRITERIA:

Patients with type 1 DM, acute complications of DM and history of acute infection or other ailments like gross CHF , inherited disorder of lipid metabolism, liver disease, endocrine diseases, tuberculosis, gout, rheumatoid arthritis, skeletal muscle injury and renal failure.

OBSERVATIONS

- Lipidogram in all three groups was compared, no significant difference in the serum Triglycerides, Total Cholestrol, LDL-C and VLDL-C levels ($p>0.05$)
- When serum HDL-C in all the three groups was compared, it was observed that HDL-C showed statistically significant decrease ($p<0.05$) in patients of DM as compared to controls.
- Serum LDL-C as well as serum Lp(a) levels showed no significant difference between patients and controls.
- Mean serum LDL levels were higher in patients by Direct method though statistically not significant and these findings were consistent with lipoprotein electrophoresis when Beta fraction was compared in patients and control.
- LDL was overestimated by Direct method when compared with Friedewald equation and serum lipoprotein electrophoresis.
- 2 patients of Group C and 1 patient of Group B and none of control Group A with Type III hyperlipoproteinemia disorder.

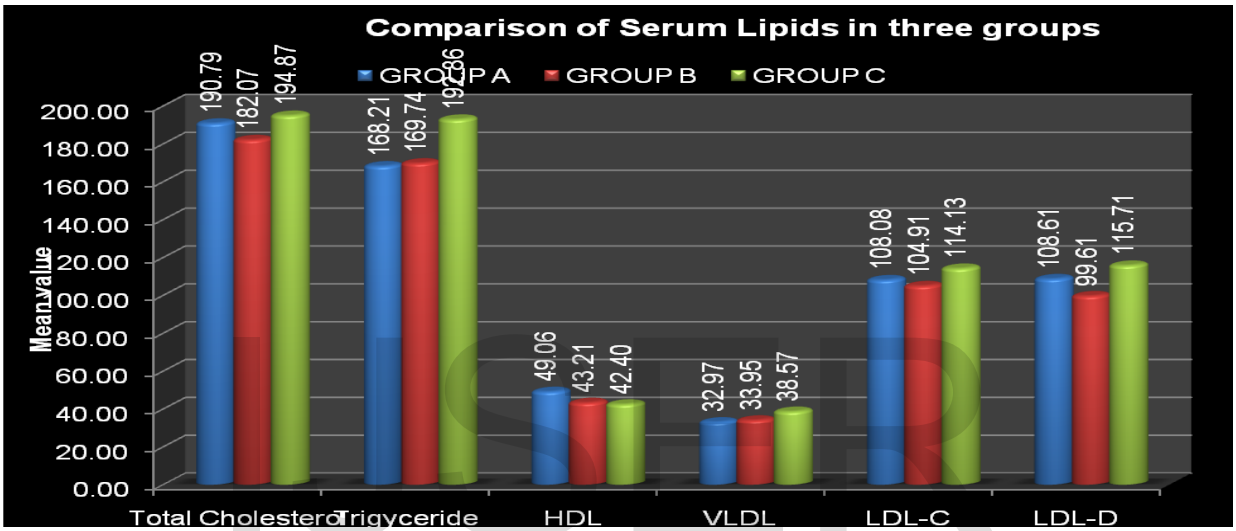

Figure 1

- Alpha lipoprotein pattern observed that percentage of serum Alpha lipoprotein showed statistically significant decrease ($p<0.05$) in patients of Type 2 DM as compared to controls.
- When the serum lipoproteins patterns on electrophoresis in Type 2 DM patients and controls were compared, it was observed that 66.6% of Group B and 63.3% of Group C showed decreased Alpha pattern as compared to 46.6% of Group A.

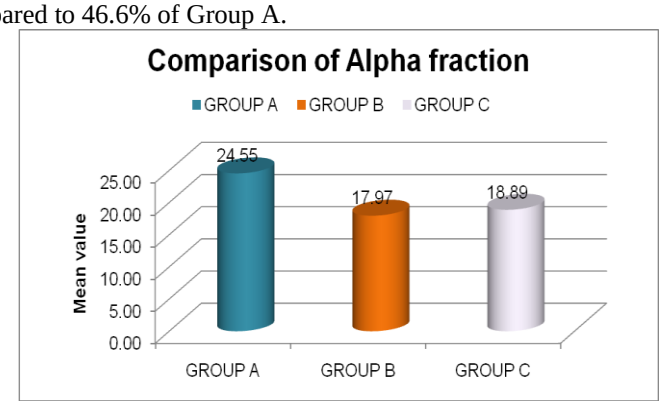

Figure 2

- Serum Pre-Beta lipoprotein was significantly increased in Group B as compared to Group A (p=0.02), Whereas no statistically significant difference was seen, when Group A and Group C were compared (p=0.250).
- Serum lipoprotein pattern on electrophoresis observed that difference in the percentage of serum Beta lipoprotein levels between the groups was statistically insignificant (p=0.681)
- It was further observed that 66.6% of Group B and 53.3% of Group C showed increased Pre-Beta pattern as compared to 23.3% of Group A.
- Increased Beta pattern was observed in 3.3% of Group B and 13.3% of Group C as compared to 6.6% of Group A subjects.

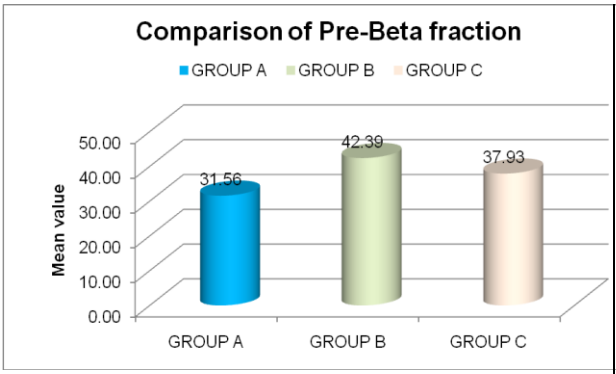

Figure 3

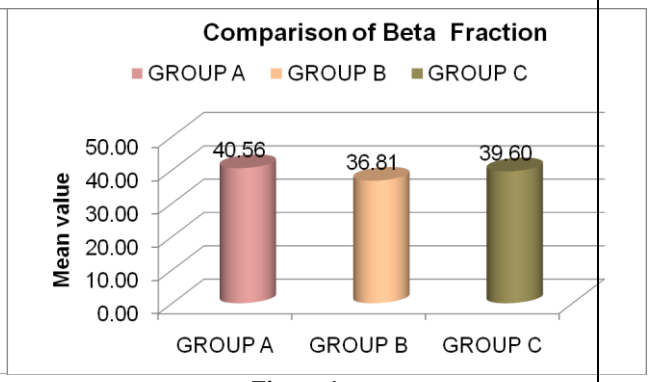

Figure 4

- Lp (a) is not statistically significant (p>0.05) when compared in present study. Although 95% Confidence interval was higher in Group C as compared to Group B, which was higher when compared to Group A.

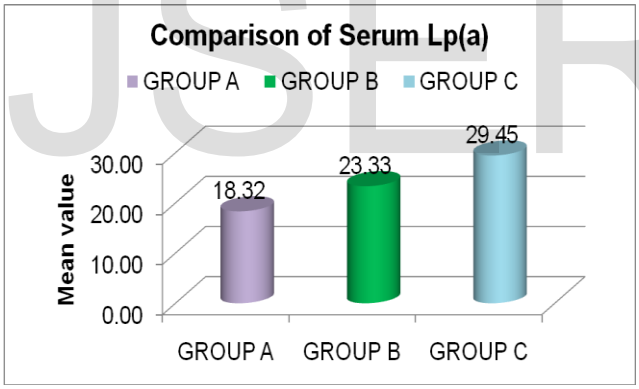

Figure 5

Table 1: OBSERVATIONS ACCORDING TO FREDRICKSON'S CLASSIFICATION OF HYPERLIPIDEMIA

TYPE	FINDING OF LIPOPROTEIN ELECTROPHORESIS	GROUP A (Non- Diabetic) N=30	GROUP B (HbA1c <7%) N=30	GROUP C (HbA1c>7%) N=30
I	Hyperchylomicronemia	03	02	03
IIa	Hyperbetalipoproteinemia	02	01	03
IIb	Mixed hyperlipidemia	00	00	01
III	Broad – beta mixed Hyperlipidemia	00	01	02
IV	Endogenous hypertriglyceridemia: (Induced by carbohydrates)	07	16	12
V	Exogenous & Endogenous hypertriglyceridemia	00	02	02
	Normal Pattern	18	08	07

Table 2: COMPARISON OF LDL-C, LDL-D AND BETA FRACTION OF LIPOPROTEINS						
TYPE	**GROUP A**		**GROUP B**		**GROUP C**	
	LDL-C & LDL-D	**Beta & LDL-D**	**LDL-C & LDL-D**	**Beta & LDL-D**	**LDL-C & LDL-D**	**Beta & LDL-D**
r value	0.911**	0.988**	0.950*	1.000**	0.952**	0.519*
p value	0.000	0.000	0.000	0.000	0.000	0.003
r value: Pearson correlation coefficient, **correlation is significant at 0.01 level, ** correlation is significant at 0.05 level						

DISCUSSION

- Lipidogram in all three groups was compared, no significant difference in the serum Triglycerides, Total Cholestrol, LDL-C and VLDL-C levels ($p>0.05$) was observed although 95% Confidence Interval for mean was slightly higher in normal healthy individuals[8].
- When serum HDL-C in all the three groups was compared, it was observed that HDL-C showed statistically significant decrease ($p<0.05$) in patients of DM as compared to controls[9].
- Serum lipoprotein pattern on electrophoresis observed that difference in the percentage of serum Beta lipoprotein levels between the groups was statistically insignificant ($p=0.681$)[10].
- Serum Pre-Beta lipoprotein was significantly increased in Group B as compared to Group A ($p=0.02$), Whereas no statistically significant difference was seen, when Group A and Group C were compared ($p=0.250$)[11.12.13].
- Alpha lipoprotein pattern observed that percentage of serum Alpha lipoprotein showed statistically significant decrease ($p<0.05$) in patients of Type 2 DM as compared to controls[10].
- Lp (a) had no statistically significance ($p>0.05$) when compared in present study. Although 95% Confidence interval was higher in Group C as compared to Group B, which was higher when compared to Group A[14].
- Nordestgaard BG suggested that LDL-C includes IDL-C and Lp(a) [15] and also in our study serum LDL-C as well as serum Lp(a) levels showed no significant difference between patients and controls.
- In the present study, mean serum LDL levels were higher in patients by Direct method though statistically not significant and these findings were consistent with lipoprotein electrophoresis when Beta fraction was compared in patients and control[16].
- Present study demonstrated that LDL was overestimated by Direct method when compared with Friedewald equation and serum lipoprotein electrophoresis. A study by Kamezaki F demonstrated that the direct measurement showed a higher rate of hypercholesterolemia prevalence than the Friedewald calculation indicated[16,18,19].
- When the serum lipoproteins patterns on electrophoresis in Type 2 DM patients and controls were compared, it was observed that 66.6% of Group B and 63.3% of Group C showed decreased Alpha pattern as compared to 46.6% of Group A[8].
- It was further observed that 66.6% of Group B and 53.3% of Group C showed increased Pre-Beta pattern as compared to 23.3% of Group A[8].
- Increased Beta pattern was observed in 3.3% of Group B and 13.3% of Group C as compared to 6.6% of Group A subjects[8].
- In our study, 2 patients of Group C and 1 patient of Group B and none of control Group A with Type III hyperlipoproteinemia disorder. Beaumount JL et al stated that lipoprotein electrophoresis can also be used in addition to other routine lipoprotein screening procedures, when more precise identification of serum lipoprotein components is needed i.e for presumptive diagnosis of Type III hyperlipoproteinemia disorder[20].

The limitations of our above study were:

(1) After complete electrophoretic run, strip cannot be preserved as bands fade away.
(2) Dye and buffer are to be prepared fresh. Shelf life is less than 7 days and has to be stored in refrigerator at 2˚C after use.
(3) Lipoprotein electrophoresis required expertise to conduct the test and interpretation of results, so to incorporate it in routine testing along with lipidogram can be a challenge.

CONCLUSION

Serum lipoprotein electrophoresis is a reliable and accurate method which can be used in clinical laboratory for screening abnormal serum lipoproteins in Type 2 DM patients and when a more presumptive diagnosis of phenotypes is needed, it could be an investigation of choice.

ABBREVIATIONS

CI	Confidence Interval
DM	Diabetes Mellitus
FBS	Fasting Blood Glucose
HDL-C	High density lipoprotein cholestrol
HbA1c	Glycosylated Haemoglobin
IDL-C	Intermediate density lipoprotein cholestrol
LDL-C	Low density lipoprotein cholesterol
Lp(a)	Lipoprotein a
LDL-D	Low density lipoprotein direct
NS	Non- Significant
Type 2 DM	Type 2 Diabetes Mellitus
TG	Triglycerides
VLDL-C	Very low density lipoprotein cholesterol
%	Percentage

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