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Original Research Article.

Study of Serum Apolipoprotein A_1 and Apolipoprotein B_{100} in Ischemic Heart Disease

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ABSTRACT

Background: Apolipoprotein A_1 (Apo A_1), the major apoprotein in HDL promotes reverse cholesterol transport producing an antiatherogenic effect in body. Apolipoprotein B_{100} (Apo B_{100}) a major apolipoprotein of LDL has atherogenic property. The aim of the study is to evaluate concentrations of Apo A_1 & Apo B_{100} in ischemic heart disease patients and healthy controls and also to establish the inter-relationships between Apo A_1 & Apo B_{100} among the patients.

Methods: A case control study carried out from October 2014 to August 2016 in the Department of Biochemistry in collaboration with Department of Medicine, Regional Institute of Medical Sciences, Imphal, Manipur. 60 ischemic heart disease (IHD) patients and 60 healthy controls were taken as the study population. The results were analysed by ELISA.

Results: The results of this study agree with the previous findings of other studies that the serum levels of Apo A_1 & Apo B_{100} are significantly altered between IHD patients and controls. However, no significant correlation has been found between these three cardiac biomarker in this study. Apolipoprotein B_{100} measurements provide useful information for IHD risk assessment.

Conclusion: The present data considers the measurement of Apo B_{100} as an important screening tool for the risk assessment of ischemic heart disease. Therefore, this study suggests the need for routine measurement of Apo A_1 & Apo B_{100} in the diagnosis of IHD and thus helps in early detection of myocardial damage which warrants timely intervention leading to lowered morbidity and mortality.

Keywords: Apolipoprotein A_1, Apolipoprotein B_{100}, Ischemic Heart Disease, ELISA.

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INTRODUCTION

Acute Myocardial Infarction (MI) is one of the major causes of morbidity and mortality in the world and atherosclerosis is the major cause of ischemic heart disease.[1] The most common cause of myocardial ischemia is atherosclerotic disease of an epicardial coronary artery (or arteries) sufficient to cause a regional reduction in myocardial blood flow and inadequate perfusion of the myocardium supplied by the involved coronary artery.[2] There are few studies on serum concentrations of apolipoprotein A_1 & apolipoprotein B_{100} and their interrelationships in IHD. Some studies showed that these individual parameters are independent and continuous risk variables for IHD and some showed that these are not independent risk factors. Hence, the present study is carried out to evaluate the relationship between apolipoprotein A_1 & apolipoprotein B_{100} in patients with IHD.

AIMS & OBJECTIVES

A) To evaluate the concentrations of Apolipoprotein A_1 (ApoA_1) & Apolipoprotein B_{100} (Apo B_{100}) in Ischemic Heart Disease patients and healthy controls.

B) To establish the inter-relationships between Apolipoprotein A_1 & Apolipoprotein B_{100} among the patients.

MATERIALS & METHODS

The study has been conducted after the approval from the institutional ethical committee. The informed consent of the patients or their relatives has been taken prior to the inclusion.

Study Design: Case control study

Study Settings: The study has been conducted in the Department of Biochemistry in collaboration with the Department

of Medicine, Regional Institute of Medical Sciences, Imphal, Manipur.

Study Duration: The study has been carried out for a period of 22 months with effect from October 2014 to August 2016.

Study Population: Consecutive patients with IHD attending RIMS, Imphal.

Subjects: A total number of 120 subjects participated in the study, of which 60 were cases and 60 were controls.

Inclusion Criteria

- All patients with already diagnosed ischemic heart disease who attended Cardiac clinic/Medicine OPD or admitted in the Medicine ward or ICCU.
- All patients above 18 yrs of age, irrespective of sex, religion and socio economic status.

Exclusion Criteria

Ischemic heart disease patients with

- Hepatic and renal disease
- Sepsis
- Malignancy
- Diabetes mellitus
- Anemia
- Hypolipidemic drugs

Collections of Blood Sample

About 6 ml of venous blood was drawn under aseptic conditions in a sterile vacutainer from selected subjects after a period of overnight fasting of 12 hours. Serum was immediately separated by centrifugation and used for analysis of Apolipoprotein A_1 & Apolipoprotein B_{100}.

Estimation of Apo A_1 & Apo B_{100}

Apo A_1 and Apo B_{100} has been estimated by using Kinesis DX's Enzyme Linked Immunosorbent assay ELISA^{PRO} KIT, manufactured by Kinesis DX Los Angeles, CA 90007, USA, as described by Nakajima K et al.[3]

Statistical Analysis: Statistical analysis was done using SPSS version-20 software. Results were reported as mean ± SD (standard deviation) for quantitative variables and number of cases along with percentages for the categorical/quantitative variables. Chi-square test, independent sample T test were applied whenever necessary. All comparisons were two-sided and the P- values of < 0.05 and < 0.01 were used as the cut-off values for significance and highly significance respectively.

Ethical Issues: Approval was sought from Research Ethics Board, RIMS, Imphal. Consent was taken before taking blood samples and confidentiality was maintained.

Table 1: Variation of the concentration of Apolipoprotein A_1 & Apolipoprotein B_{100} among different age groups in case

	31-40 years	41-50 years	51-60 years	>60 years	ANOVA significance
ApoA_1 (µg/ml)	1371.42±447.95 µg/ml	1211.76±706.11 µg/ml	1199.03±718.82 µg/ml	1282.43±1105.05 µg/ml	f=0.19 p=0.90
Apo B_{100} (mg/ml)	1.05±0.72 mg/ml	1.17±0.87 mg/ml	1.48±1.12 mg/ml	1.57±0.88 mg/ml	f=1.41 p=0.24

Table 2: Variation of the concentration of Apolipoprotein A_1 & Apolipoprotein B_{100} among both sex groups in case

	Male(32)	Female(28)	Significance t test
Apo A_1 (µg/m)l	1265.75±676.63 µg/ml	1217.02±1024.39 µg/ml	t=0.314 p=0.754
Apo B_{100}(mg/ml)	1.29±0.91 mg/ml	1.61±1.06 mg/ml	t=-1.754

Table 3: Distribution of the respondents by serum concentrations of Apo A_1 & Apo B_{100}

Variable	Mean ± SD
Ischemic heart disease	
Apo A_1	748.3± 821.7 µg/ml
Apo B_{100}	2.1 ± 0.8 mg/ml
Healthy controls	
Apo A_1	1745.0 ± 444.5 µg/ml
Apo B_{100}	0.6 ± 0.3 mg/ml

Table 4: Correlation between Apo A_1 & Apo B_{100}

Variable	Mean ± SD	Unpaired t-test
Apo A_1 (µg/ml)		
Case	748.3 ± 821.7 µg/ml	t=8.2
Control	1745 ± 444.5 µg/ml	p<0.001
Apo B_{100} (mg/ml)		
Case	2.1 ± 0.8 mg/ml	t=12.33
Control	0.6 ± 0.3 mg/ml	p-<0.001

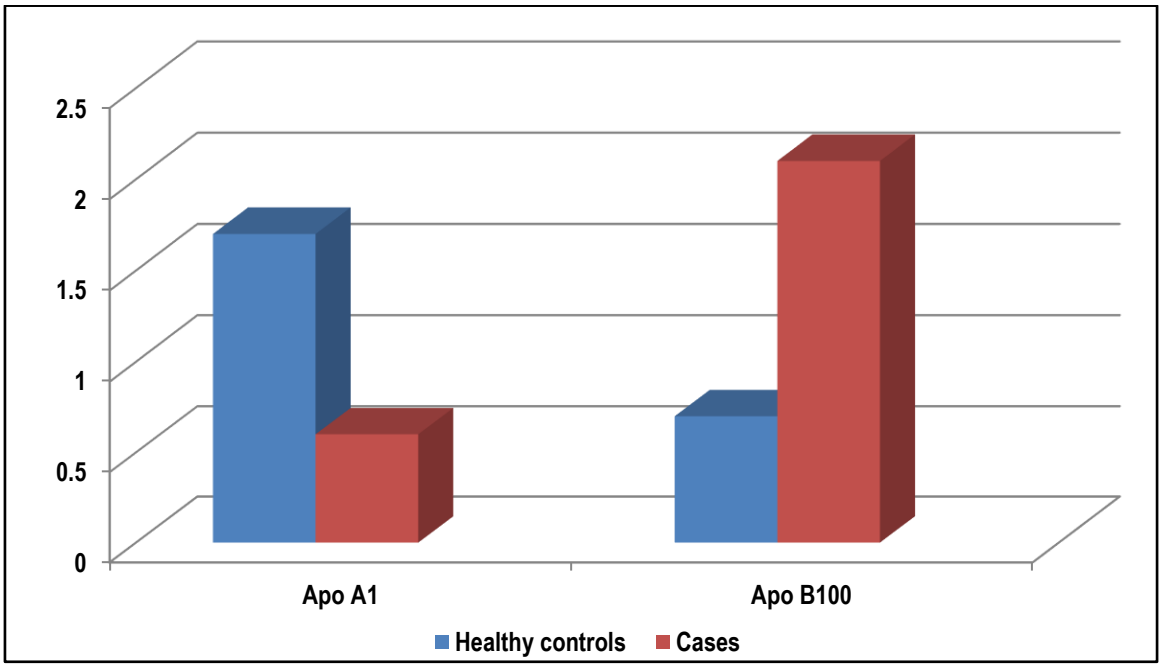

Figure 1: Bar diagram showing serum concentrations of Apo A1 & Apo B100 in Healthy Controls and cases.

RESULTS

Table 1 shows variation of the concentration of Apolipoprotein A_1 & Apolipoprotein B_{100} among different age groups where Apo A_1 concentration gradually decreases where concentration of Apo B_{100} has been gradually increases with increment of age. Comparison between different age groups among cases by ANOVA test was statistically insignificant ($p>0.05$).

Table 2 shows variation of the concentration of Apolipoprotein A_1 & Apolipoprotein B_{100} among both male and female groups in case, where ApoA_1 has been found insignificantly higher among males(32) and Apo B100 was insignificantly higher among females(28) ($p>0.05$).

Table 3 & Figure 1 depicts that mean with standard deviation of Apo A_1 & Apo B_{100} were 748.3± 821.7 µg/ml & 2.1 ± 0.8 mg/ml respectively for Ischemic heart disease and for healthy controls 1745.0 ± 444.5 µg/ml & 0.6 ± 0.3 mg/ml.

Table 4 significantly shows that on comparing various level of Apo A_1 & Apo B_{100} of cases with the controls, for Apo A_1 cases had lower level than controls but for Apo B_{100} the cases had higher values than controls. All the findings were statistically significant ($p<0.05$)

DISCUSSION

The present study was undertaken to determine the association of various risk factor variables to detect the presence of IHD. The details of the study group are described in the results. In the first part of the results, an attempt has been made to find out levels of Apolipoprotein A_1 & Apolipoprotein B_{100} among different age groups. Table 1 shows variation of the concentration of Apolipoprotein A_1 & Apolipoprotein B_{100} among different age groups where Apo A_1 concentration gradually decreases & concentration of Apo B_{100} gradually increases with increment of age. Comparison between different age groups among cases by ANOVA test was statistically insignificant ($p>0.05$). This finding correlated with study by Talmud PJ et al who compared the pairwise combinations of total cholesterol, TGL, apolipoprotein B, HDL-C, LDL-C and apolipoprotein A_1, apolipoprotein B_{100} and hsCRP on CHD risk prediction in middle -aged men and younger age groups and concluded aforementioned markers significantly altered with increment of age so that the combined evaluation of Apolipoprotein B with TGs provides useful diagnostic criteria for CHD.[4] Ariyo AA et al observed in older U.S. adults that all Apolipoproteins play an important part in atherothrombogenesis and predicting increased risk of vascular disease in older men.[5]

Table 2 shows variation among both male and female groups in cases, where ApoA_1 has been found insignificantly higher among males(32),and Apo B_{100} has been found insignificantly higher among females(28) ($p>0.05$). This study is in favour of Brown G et al who reported that regression of CAD as a result of intensive lipid lowering therapy in men with high levels of apolipoprotein B and inversely proportional to high level of Apolipoprotein in both sex groups.[6] Kamstrup PR et al reported that extreme apolipoprotein levels predict a 3 to 4 fold increase in risk of MI in the general population and absolute 10-year risks of 20% and 35% in high risk women and men and male have a higher chance of having IHD as compared to female.[7]

Ischemic heart disease is one of the important causes of morbidity and mortality in most countries of the world. The debate on the value of lipids as a predictive risk factor for atherogenesis has centered for many years on total cholesterol, triglycerides and low density lipoprotein. Recently the interest has been focused on role of Apolipoprotein A_1 & Apolipoprotein B_{100} and other inflammatory marker in atherogenesis.

In the Table 4, we see that the mean serum concentration of Apolipoprotein A_1 was lower in IHD subjects when compared to controls suggesting a negative association to ischemic heart disease. These results were in accordance with the studies of Sherpa LY et al conducted in Lhasa Tibet.[8] The apolipoprotein B_{100} concentration was increased in patients with ischemic heart disease when compared to age matched healthy normal individuals. This study showed a positive association between ischemic heart disease and apolipoprotein B_{100} level and the results were in agreement with study of Miremadi S et al which

was conducted to examine whether measurement of a single marker (apo B_{100}) led to the same categorization of risk as the traditional five indices (lipid profile).[9] Our findings are supported by another study by Ansari KMH et al who concluded that insulin, APO-A1, APO-B_{100}, and total cholesterol measurement is a good case for defining the severity of coronary artery involvement, while high-density lipoprotein, low-density lipoprotein, and triglyceride are not important risk-factors.[10] Another study done by Ryoo JH et al found ApoB_{100} to be independently related to the risk of CHD.[11]

CONCLUSION

The results of the current study support the concept that the levels of Apolipoprotein A_1, Apolipoprotein B_{100} and hsCRP are strongly related to ischemic heart disease in addition to the conventional lipid profile. Apolipoprotein B_{100} measurements provide useful information for CHD risk assessment. Our findings support the consideration of the measurement of serum Apolipoprotein B_{100} as a screening tool for the risk of ischemic heart disease.

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