The present study is an observational case control study, comprising 160 subjects. Out of these 110 were in study group and 50 were in control group.

Out of 160 subjects, 97 were male (70.1%), 63 females (29.9%), with 63 between 30-40 (reproductive age group) and 22 from 51 to 60 years. In other studies similar findings were noted. Among the three Asian populations, Chinese, Malay, and Indian, the highest age-standardized incidence rates in both sexes are in Indians. The first MI attack occurs in 4.4% of Asian women and 9.7% of men at age less than 40 years, which is 2-3.5 fold higher than in the West European population and is third highest of all the regions, studied worldwide. These studies carried out in India and other places suggest that Asians in general and Indians in particular are at increased risk of CHD at a younger age (<40 years), irrespective of whether they have migrated to other countries or are resident Asians. Reports on CHD in Indians from different parts of the world have shown that Asian Indians are at 3–4 times higher risk of CHD than white Americans, 6 times higher than Chinese, and 20 times higher than Japanese.

Overall, majority of the patients were from urban areas, with maximum number of patients (72 in study group) having sedentary life style. Various studies showed that sedentary life style could be an independent risk factor for CHD. A low level of physical activity has been noted to be a significant risk factor for premature CHD in Indians.

In the present study, out of total 160 patients, 92 were non-vegetarian with predominance of male patients (79.3%). It has been observed earlier that a diet rich in plant food is associated with a lower risk of cardiovascular diseases. A large scale systematic review of the association between diet and CHD concluded that the beneficial substances in a vegetarian diet have a prominent role in reducing risk of CHD.

A total of 77 (48.1%) patients had family history of diabetes mellitus and 52 patients (47.3%) had family history of CHD. Studies from the past showed that diabetes is a powerful risk factor for CHD. Up to 75–80% of adult diabetic patients die of cardiovascular diseases, and 75% of these deaths are caused by CHD. There are large numbers of CHD patients, who succeed in meeting their target “LDL-C goal” as per suggested by ATP III guidelines, but still develop complications from atherosclerotic vascular disease and suffer from cardiovascular events. These patients bear the burden
Discussion

of having residual risk not identified by using traditional metabolic and cardiovascular markers like LDL-C. Because of these reasons clinicians are looking forward to new and more reliable risk markers to predict CHD in these patients. Epidemiologic research and clinical event trials have provided abundant evidence that, in comparison with the traditional risk markers the new and emerging risk markers like non-HDL-C, apoB, apoA-1 and Lp(a) have an important role in the initial assessment and ongoing monitoring of CHD for coronary events and stroke.17, 30, 181

Whayne TF et al suggested that measurement of apoB offer important predictive value for coronary artery disease, especially at lower levels of plasma cholesterol.182 The strong predictive value of apoB was confirmed in the apolipoprotein-related mortality risk (AMORIS) study that included more than 175000 individuals. This study has shown that high levels of apoB are strongly related to increased CHD risk. The AMORIS trial also demonstrated that apoB was a better risk predictor than LDL-C, even in individuals with concentrations of LDL-C below the median. This was a significant finding because up to 50% of patients had normal cholesterol (<200 mg/dl), yet a large proportion will have elevated apoB concentrations and therefore, residual risk. 100 Standardization of apoB assay has largely been successful due to availability of suitable reference materials and support from the International Federation of Clinical Chemistry (IFCC). 183

The American Diabetes Association/American College of Cardiology (ADA/ACC) Consensus Conference Report and the Canadian Cardiovascular Society have suggested apoB levels for treatment of dyslipidemia and for prediction and prevention of cardiovascular disease.30 The ADA/ACC consensus report recommends an apoB goal of 80 mg/dl for patients with either established cardiovascular disease or diabetes with one risk factor. In patients without cardiovascular disease but with cardiometabolic risk factors, the ADA/ACC recommends an apoB goal of 90 mg/dl. For patients with coronary heart disease, the Canadian Cardiovascular Society recommends an apoB goal of 80 mg/dl. Our findings suggest a cut off value of 89 mg/dl to determine CHD risk.

In the present study, apoB was observed as the strong emerging risk factor. The value of apolipoprotein B was significantly (p<0.001) elevated in study group as compared to control group (Table 5.13). 44 (40%) patients were found to have normal apoB (<98 mg/dl) and 66 (60%) patients were found to have abnormal (>98 mg/dl) values. When apoB was compared with varying levels (normal and elevated) of Lp(a), HDL-C, TG, TC
and apoA-1, it was observed that apoB had highly significant (p<0.001) correlation with Lp(a) while, it had significant correlation with TG, and TC. Apo B could not correlate with normal as well as elevated levels of LDL, non-HDL-C and apoA-1. Similar kind of results was observed by de Bruin et al.\textsuperscript{184} in their study.

Non-HDL-C was introduced as another means to refine risk estimation beyond LDL-C from Friedewald’s formula in the presence of raised triglycerides (TG) levels (≥200 mg/dl), since associated changes in VLDL-TG/VLDL-C ratio may lead to LDL-C under calculation. As it actually estimates the level of all apoB-carrying lipoproteins, non-HDL-C may represent a simple and inexpensive surrogate to apoB measurement, especially in selected patients groups, such as hypertriglyceridemic patients.\textsuperscript{27, 30, 185, 186}

There are several advantages of non-HDL cholesterol measurement over apolipoproteins. First, it makes no assumption about the relationship between VLDL cholesterol and triglycerides, this relationship can be altered, leading to falsely low LDL values as calculated by the Friedewald formula, especially in conjunction with elevated triglyceride levels. Secondly, non-HDL cholesterol includes an assessment of all apolipoprotein B containing lipoproteins considered to be atherogenic, i.e., VLDL, intermediate-density lipoprotein (IDL), and LDL, and even lipoprotein (a). Finally, non-HDL cholesterol has several practical advantages in a clinical setting, including the ability to be assessed in patients with triglyceride level > 400 mg/dl and in patients who are not fasting.\textsuperscript{187, 188}

According to the NCEP guidelines the optimal level for non-HDL-C should be <130 mg/dl.\textsuperscript{188} In the present study cutoff value for non-HDL-C observed was 124.5 mg/dl and it was found to be significantly elevated (p>0.001) in study group as compared with the control group (Table 5.11), out of total patients 32 (29.1%) were found to have normal (<130 mg/dl) and 78 (70.9%) patients were found to have abnormal (>130 mg/dl) levels of non-HDL-C. When compared with TC, LDL-C, HDL-C, and TG it was observed that non-HDL-C had highly significant (p<0.001) correlation with varying values (normal and elevated) of TC (<200 and >200 mg/dl), LDL-C (<100 and >100 mg/dl), HDL-C (≤40 and >40 mg/dl), and TG (<150 and >150 mg/dl).

Similar kinds of results were also observed in the previous studies. Findings from the Lipid Research Clinics Program Follow-Up Study, in which a total of 4,462 men and women were followed for 19 years, showed that non-HDL cholesterol emerged as a
somewhat better predictor of CVD mortality than LDL. In the Systolic Hypertension in the Elderly Program (SHEP), a study of elderly, primarily white, non-HDL cholesterol was assessed along with serum cholesterol, triglycerides, LDL cholesterol, and HDL cholesterol as a predictor of cardiovascular events in 4,736 participants. During an average 4.5 years of follow-up, non-HDL cholesterol was shown to be a predictor of cardiovascular events in multivariate analysis in the total population. In the present study also, non-HDL-C was found to be significantly elevated in study group as compared with the control population, 32 (29.1%) patients were found to have normal non-HDL-C (<130 mg/dl) and 78 (70.9%) patients were have abnormal (>130 mg/dl) levels of non-HDL-C.

Elevated levels of Lp(a) have long been known to be associated with premature CHD. In cross-sectional analysis of the Lipid Research Clinics Coronary Primary Prevention Trial and the Framingham Heart Study, it was shown that elevated levels of Lp(a), whether assessed by immunoassay or electrophoresis, were significantly associated with CHD risk. Tetsuya Ohira et al found that increased levels of Lp(a) were associated positively and independently with the incidence of ischemic stroke in black and white women. Danesh et al, in a meta-analysis including the prospective studies published before 2000, concluded that there was a clear association between lipoprotein (a) and CHD in the general population. Hiraga et al, demonstrated that lipoprotein (a) was an independent risk factor for CVD in Japanese type 2 diabetic patients.

The results of various studies showed significant correlation between levels of Lp(a) and CHD. Upper limits of normal Lp(a) are not defined in Indian population. In Caucasian population Lp(a) 30 mg/dl is considered as upper limit of normal Lp(a) levels. In Indian population Enas et al. suggested Lp(a) 20 mg/dl as upper limits of normal Lp(a). A previous study from Indian region, observed the mean of 16.04 mg/dl and suggested the cut off level of 25mg/dl for Lp(a) to determine the risk of CHD. In consistent with these findings our study suggested a cut-off level of 20 mg/dl for Lp(a) to determine the risk of CHD.

In the present study, lipoprotein (a) was observed as an important and an independent risk factor. The levels were significantly elevated (p<0.001) in the study group as compared with control group (Table 5.15). Out of total patients, 26 (23.6%) were found to have normal Lp(a) (<20 mg/dl) and 84 (76.4%) patients were found to have abnormal (>20
mg/dl) levels. When Lp(a) was compared with apoB, non-HDL-C and apoA-1, it was found that apoB had highly significant (p<0.001) correlation with normal as well as elevated levels of Lp(a), while non-HDL-C had significant correlation and apoA-1 could not correlate with varying levels of Lp(a). These observations suggest that in addition to conventional lipid profile, estimation of Lp(a) can prove to be a valuable tool in risk assessment of population in general and management of disease in particular and should be routinely screened.

We observed that lipid and lipoprotein ratios i.e TC/HDL-C, LDL-C/HDL-C, TG/HDL-C, apoB/apoA-1 and non-HDL-C/HDL-C levels were statistically significant in study group but they could not correlate with age, diet, locality, and F/H of DM or CHD.

Various studies found that TC/HDL is a better predictor of CHD risk in comparison with LDL-C. TC/HDL-C ratio was a useful and simple index of CHD risk in men in the Quebec Cardiovascular Study. It is proposed that the ability of this ratio to predict risk is explained by the fact that it is a relevant cumulative marker of the cluster of metabolic abnormalities found in individuals with high TG–low HDL-C dyslipidemia. Similarly, Wang TD et al, Nair D et al and Benoit J. Arsenault et al concluded in their respective studies the ratio of TC/HDL was significantly associated with the increased risk of CHD.

Benoit J. Arsenault et al in their study showed that patients with TC/HDL-C ratio levels >5 mg/dl were at increased CHD risk. Similarly, Wang TD et al showed that high total cholesterol/HDL cholesterol ratios (>5mg/dl) had a 2.5-fold higher incidence of CHD than those with similar LDL cholesterol levels. Our findings suggest a cut-off value of 3.84mg/dl to determine the risk of CHD. In the present study TC/HDL was found to be significantly (p<0.01) elevated in the study group as compared with the control group, 52 (47.3%) were found to have normal (<4.50 mg/dl) and 58 (52.7%) were found to have abnormal (>4.50mg/dl)TC/HDL-C levels (Table 5.26). When compared with varying levels (normal and elevated) of TC, LDL-C, HDL-C, TG, non-HDL-C, apoB, apoA-1 and Lp(a), it was observed that TC/HDL had highly significant correlation with varying levels of TG, non-HDL-C and it could not correlate with the varying levels of apoB and apoA-1.
Several studies have demonstrated that the LDL-C/HDL-C ratio is an excellent predictor of risk of CHD.\textsuperscript{60,139,140} In the Helsinki Study, a clinical trial with a 5-year follow-up, involving more than 4000 middle-aged men with hyperlipidemia, the LDL-Cholesterol/HDL-C ratio had a superior prognostic value compared with isolated values of LDL-C and HDL-Cholesterol. The predictive ability of this ratio was particularly strong in patients with concomitant elevation of triglycerides. Similarly, Stampfer et al.,\textsuperscript{139} Manninen et al.,\textsuperscript{60} Bruce Kinosian et al.,\textsuperscript{56} and Cullen et al.\textsuperscript{140} concluded that LDL-C/HDL-C ratio had a greater correlation with cardiovascular disease and are therefore is the better predictor of cardiovascular disease than simple lipid parameters. In the present study LDL-C/HDL-C was found to be significantly (p<0.001) elevated in the study group as compared with the control group (Table 5.25), 49 (44.5%) patients were found to have normal (<3 mg/dl) and 61 (55.5%) patients had abnormal (>3 mg/dl) levels of LDL/HDL-C and our findings suggest a cut off value of 2.36mg/dl to determine CHD risk.

Many clinicians and researchers have found TG/HDL ratio to be one of the better predictors of heart disease.\textsuperscript{63,139,144} Research has shown that people with the highest ratio of triglycerides to HDL cholesterol has 16 times the risk of heart attack as those with the lowest ratio of triglycerides to HDL. Protasio L et al in their study showed that the ratio of TG/HDL was robustly associated with CV diseases.\textsuperscript{147} Elevation in the ratio of TG to HDL-C was the single most powerful predictor of extensive coronary heart disease among all the lipid variables examined. Similarly, Hadaegh F et al,\textsuperscript{148} Quijada Z et al,\textsuperscript{53} Roa Barrios M et al,\textsuperscript{150} and Procolo Di Bonito et al\textsuperscript{151} in their respective studies concluded that TG/HDL-C ratio was a better risk marker for cardiovascular diseases. Li Tian et al and Martin R et al in their respective studies showed that elevated level of TG/HDL-C ratio (>2.5 and >3.5 for women and men) was associated with increased risk of CHD.\textsuperscript{199,200} Our findings suggest a cut-off value of 2.68 mg/dl for TG/HDL-C ratio to determine the risk of CHD.

In the present study, TG/HDL-C ratio was found to be significantly (p<0.001) elevated in study group as compared with control group (Table 5.25), 18 (16.4%) patients were found to have normal (<2.5 mg/dl) and 92 (83.6%) patients had abnormal (>2.5 mg/dl) TG/HDL-C levels.

The ratio of apoB/apo A-1 has also been proposed as a superior measure of the ratio of proatherogenic (i.e. “bad”) cholesterol to anti-atherogenic (i.e. “good”) cholesterol. This
ratio may be a more accurate measure of this concept, compared to the more common total cholesterol/high-density lipoprotein cholesterol (TC/HDL-C) ratio. A number of epidemiologic studies have reported that the apoB/apo A-1 ratio is superior to other ratios, such as TC/HDL-C, or low density lipoprotein cholesterol (LDL)/HDL-C.\textsuperscript{161,157,201} Kappelle et al\textsuperscript{202} used data from the prospective PREVEND cohort to evaluate the predictive value of the apoB/apoA-1 ratio independent of other traditional risk factors, including albuminuria and C-reactive protein (CRP). Among 6,948 individuals without previous heart disease and who were not on lipid-lowering drugs, the adjusted hazard ratio for a high apoB/apoA-1 ratio was 1.37 (95\% CI: 1.26-1.48). This hazard ratio was not significantly different from the total cholesterol/HDL-C ratio of 1.24 (95\% CI: 1.18-1.29), and was not significantly changed after further adjustment for triglycerides. Some studies have tested the use of apo \textit{B} in a multivariate risk prediction model in which both traditional risk factors and apolipoprotein measures were included as potential predictors. Ridker and co-workers\textsuperscript{203} published the Reynolds Risk Score, based on data from 24,558 initially healthy women enrolled in the Women’s Health Study and followed up for a median of 10.2 years. A total of 35 potential predictors of cardiovascular disease were considered as potential predictors, and 2 final prediction models were derived. The first model was the best fitting model statistically, and included both apo \textit{B} and the apoB/apoA-1 ratio as 2 of 9 final predictors. The second model, called the “clinically simplified model,” substituted LDL-C for apoB and total/high density lipoprotein (HDL) cholesterol for apoB/apo A-1. The authors developed this simplified model “for the purpose of clinical application and efficiency” and justified replacing the apo-\textit{B} and apo \textit{B}/apo A-1 measures as a result of their high correlation with traditional lipid measures (\(r=0.87\) and 0.80, respectively). Previous reports have shown that apoB/apoA-1 ratio for men and women respectively <0.7 and <0.6 is associated with low risk for cardiovascular disease.\textsuperscript{100,157,200} According to Walldius et al apoB/apoA-1 ratio within 0.3-0.6 for women was considered as low risk and >0.8 as high risk of myocardial infarction.\textsuperscript{100} Our findings suggest a cut off level of 0.70mg/dl to determine CHD risk.

In the present study apoB/apoA-1 ratio was found to be significantly (\(p<0.001\)) elevated in study group as compared with control group (Table 5.25); 39 (35.5\%) patients were found to have normal (<0.70 mg/dl) and 71 (64.5\%) patients were have abnormal (>0.70 mg/dl) apoB/apoA-1 levels. When compared with varying levels (normal and elevated) of
TC, LDL-C, HDL-C, TG, non-HDL-C, apoB, apo- A1, and Lp(a), it was observed that apoB/apoA-1 had highly significant correlation with varying values of apoB and apoA-1 while it had significant correlation with varying values of HDL-C, and it could not correlate with varying levels of TC, LDL-C, TG, and non-HDL-C.

In the present study non-HDL-C/HDL-C was also observed as an emerging and important risk predictor of CHD. However, the significance of non-HDL cholesterol/HDL cholesterol (non-HDL-C/HDL-C) ratio, as another cardiovascular risk predictor in Indian population, has not been evaluated so far to best of our knowledge. Eliasson B et al in their study concluded that lower levels of non-HDL-C/HDL-C are a better risk marker for CHD than LDL-cholesterol. 75 Similarly, Kim SW et al observed in their study that the non-HDL-C/HDL-C ratio is a better marker for identifying insulin resistance and metabolic syndrome.76

The present study is perhaps first Indian study to evaluate the efficacy of this parameter in the prediction of CHD. In the present study non-HDL-C/HDL-C was found to be significantly (p<0.001) elevated in study group as compared with control group (Table 5.25); 26 (23.6%) patients were found to have normal (<3 mg/dl) and 84 (76.4%) patients were found to have abnormal (>3 mg/dl) non-HDL-C/HDL levels. Our study suggests a cut off value of 2.85 to determine CHD risk.

When non-HDL-C/HDL-C was compared with varying (elevated and normal) levels of LDL-C, TC, HDL-C, TG, non-HDL-C, apoB, apoA-1, and Lp(a), it was observed that non-HDL-C/HDL-C had highly significant (p<0.001) correlation with varying levels (normal and elevated) of LDL-C, TC, TG and non-HDL-C, had significant (p<0.05) correlation with HDL-C and Lp(a) while it could not correlate with varying levels (normal and elevated) of apoB and ApoA-1.

In conclusion, although LDL-C is a well founded target, emerging findings suggest that it has become a suboptimal marker of risk for a number of reasons 111, 204. With the intent of assessing complete lipid atherogenic risk burden—rather than a partial one, such as LDL-C—the ideal parameter is one that accounts for all atherogenic cholesterol particles, including LDL-C, Lp(a), intermediate-density lipoprotein cholesterol, chylomicron remnants, and VLDL-C 205, 206. The dynamic flux of lipoproteins between subtypes under direction of LPL and cholesterol ester transfer protein (CETP) makes direct assessment of
total atherogenic burden a challenge, which is significantly improved by apoB and non-HDL-C \cite{205, 207}. Apolipoprotein B is able to directly measure the aggregate number of all atherogenic lipoproteins because each atherogenic particle contains 1 apoB\textsubscript{100} molecule.

Non-HDL-C is an established secondary target of therapy per the NCEP ATP III guidelines that remains underutilized in the clinical setting. \cite{208} With conventional analysis, non-HDL-C is able to quantify total atherogenic burden by measuring the aggregate amount of “cholesterol” in all contributive particles. Non-HDL-C is a quick and simple calculation of TC minus HDL-C (TC-HDL-C), and can be obtained in the non-fasting state without affecting results.

The predictive value of apoB and non-HDL-C has been a topic of debate for decades. It is well-recognized that apoB and non-HDL-C are closely related metabolically, yet there is ongoing discussion as to whether one should be measured preferentially over the other, with some considering apoB as a choice proatherogenic index in patients with cardiometabolic risk associated with atherogenic dyslipidemia. Multiple epidemiological and clinical trials support apoB and LDL particle number as the superior marker for cardiovascular risk prediction when compared to non-HDL-C and LDL-C.\cite{13, 182, 186} The INTERHEART study was a large international standardized case-control study of acute myocardial infarction involving more than 12,000 subjects, over 14,000 age- and sex-matched controls, and a variety of ethnic groups. ApoB had the highest odds ratio for prediction of vascular disease and emerged as the superior marker across all ethnic groups.\cite{31} Charles R. Harper et al (2010) found in their study that apolipoprotein B is superior than LDL-C and suggested that emphasis should be given on the use of apolipoproteins as a risk marker for better prediction of CHD.\cite{105} Ballantyne CM et al in their study showed that total serum apoB is a strong predictor for severity of coronary atherosclerosis and CAD events, and non-HDL-C is highly correlated with apoB levels. However, Keys et al, Pocock et al, Menotti et al, and Bos et al in their respective studies showed that non-HDL is better predictor of CHD.\cite{209-211}

Thus, to evaluate and compare apoB and non-HDL-C and to find out the superiority of one over the other, we compared direct LDL, calculated LDL, non-HDL-C, and apoB with normal as well as elevated levels of TG and observed that non-HDL-C had highly significant correlation (p<0.001) with normal as well as elevated levels of TG. ApoB and Direct LDL also had significant correlation (p<0.05) but non-significant results were
found for Calculated LDL (Table 5.35). Receiver operating characteristic curve was commuted to access the utility and to compare the predictive values of non-HDL-C, apoB, and LDL.

The present study confirms that non-HDL-C is equivalent to apoB on the basis of ROC curve analysis, which is in agreement with Stanley S. Levinson\textsuperscript{212} who found little difference between apoB and non-HDL-C in discriminating CAD. Sondermeijer et al\textsuperscript{213} also concluded recently that non-HDL-C and apoB were comparable in their ability to predict risk of future CHD.

In conclusion, our study has found that there is no difference between apoB and non-HDL-C in predicting cardiovascular risk. However, we would suggest the use of non-HDL-C as an initial screen for coronary risk particularly in Indian population. India being a developing country, people would not be burdened with additional cost of apoB estimation. Besides, non-HDL-C can easily be calculated from routine lipid panel in minimum time. In fact, Ramjee et al\textsuperscript{214} recently suggested non-HDL-C to be a marker of choice when compared to apoB.

Thus, non HDL-C should be added in the routine lipid profile panel especially for the screening and prediction of risk of CHD. Screening for CHD in the young population may help to improve prognosis by detecting subclinical disease, although more studies are necessary to establish reference limits. Additional research must also focus on treatment concerns.