

## Research Article

**Are lipid ratios better indices for evaluating hyperlipidemia? A prospective observational study from a rural region of North Kerala**

Simi T. K., Rebecca Abraham, Sangeetha V. Joice

Department of Biochemistry, Malabar Medical College Hospital and Research Centre, Ulliyeri, Kozhikode, Kerala, India

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Corresponding author: **Rebecca Abraham**. Email: gracefieldra@gmail.com**ABSTRACT**

**Introduction and Aim:** Historically lipid profile has been used to evaluate hyperlipidemia. More recently lipid ratios have been used particularly in studies from the west. The aim of this study is to evaluate dyslipidemia using both the conventional lipid profile and lipid ratios and to assess its relationship with fatty liver disease.

**Materials and Methods:** Fasting lipid profile, lipid ratios such as the Castelli Risk Index-I (CR-I TC/HDL), Castelli Risk Index II (CR-II LDL/HDL), atherogenic coefficient (non-HDL/HDL) were assessed in our study population of 150 subjects. Any changes noted in serum lipids were correlated with ultrasonography of the liver.

**Results:** Significant changes in lipid ratios such as CR-II was noted in the age group 50-59 years and was statistically significant ( $p=0.053$ ). CR-I, CR-II and AC indicated a cardiovascular risk prevalence of 22.6%, 36.7% and 50.7% respectively. The two ratios which were significantly altered across the various subtypes of fatty liver were LDL/HDL ( $p=0.018$ ) and non-HDL/HDL ( $p=0.00$ ).

**Conclusion:** This study emphasizes the importance of lipid ratios in early screening.

**Keywords:** Atherogenic coefficient; Castelli risk index; dyslipidemia; fatty liver.

**INTRODUCTION**

In India, there has been a steady increase in the prevalence of cardiovascular disease (CVD) over the last 20 years. Of concern is that 24% of death occurred amongst adults in the age group 25-69 years (1). The average age of onset was found to be lower amongst Indians as compared to other populations. Likely causes for the increase in CVD events include lifestyle changes and dietary practices associated with urbanization or economic development (2). Both sex-dependent differences and differences in rural and the urban communities have been reported in India (3).

When assessing dyslipidemia, the typical lipid profile includes measurements of triglycerides (TG), low-density lipoprotein-cholesterol (LDL-C), high-density lipoprotein cholesterol (HDL-C), and total cholesterol. However, LDL-C is typically given the greatest attention among these components (4). However other studies have demonstrated the significance of lipid ratios in predicting the risk of cardiovascular events (5). Two crucial factors that serve as indicators of cardiovascular risk are the Castelli Risk Index I, which is the total cholesterol to high-density lipoprotein cholesterol (HDL-C) ratio, and the Castelli Risk Index II, which is the low-density lipoprotein cholesterol (LDL-C) to HDL-C ratio (4). Recent studies have shown the importance of non-HDL-cholesterol (non-HDL-C) in the management of dyslipidemias (6).

Dyslipidemia has been closely associated with fatty liver and is recognized as a risk factor for non-alcoholic fatty liver disease (NAFLD; 7). The objective of this study was to examine dyslipidemia

using both the conventional lipid profile and lipid ratios, and to investigate its association with fatty liver disease.

**MATERIALS AND METHODS**

This is a prospective observational study. The study group consisted of subjects visiting the outpatient department of Malabar Medical College and Research Centre for a routine health check-up. The duration of the study was from July 2017 to December 2018. Patients with acute illness or features of organ failure, known cases of diabetes /or dyslipidaemia on medication and those who did not come fasting or declined consent were excluded from the study.

Informed consent was taken from the subjects and the study was cleared by the Scientific and Ethical Committees of the Institution. A detailed questionnaire was submitted to 150 subjects. Data on demographic characteristics, medical history, were recorded in the questionnaire. All study participants underwent an overnight fast before blood samples were collected to determine their lipid profile and blood sugar levels. Five ml of blood was collected by venipuncture from the antecubital vein into sterile plain vials for assessing the lipid profile and into a blood sugar vial containing the anticoagulant sodium fluoride for estimating blood sugar. The assay parameters were analysed on the Fully Automated Analyser (Erba Mannheim –EM360).

The measurement of serum cholesterol was performed using the Cholesterol oxidase/peroxidase method, also known as CHOD-PAP [8], serum triglycerides by

Glycerol Phosphate oxidase phenol-4-amino antipyrine (GPO/PAP) method (9) and direct enzymatic method was used for HDL – C (10). LDL – C level was calculated using Friedwald’s formula ie., LDL - Cholesterol = Total Cholesterol - (Triglyceride / 5+ HDL) (11). A straightforward formula is used to calculate the concentration of non-HDL-C, which is non-HDL-C = TC - HDL-C (6). As per NCEP guidelines, the cut-off values for hypercholesterolemia and hypertriglyceridemia are  $\geq 200$  mg/dl and  $\geq 150$  mg/dl respectively. Low HDL cholesterol for men is  $< 40$  mg/dl and is  $< 50$  mg/dl for women and the cut off value for non-HDL-cholesterol for high risk CVD is  $> 160$  mg/dl .Lipid ratios such as the Castelli’s risk index –I (CRI-I-TC/HDL), CRI-II (LDL/HDL), and Atherogenic Coefficient(AC) (non-HDL/HDL) are also diagnostic alternatives in predicting the risk of developing cardiovascular events and were obtained by calculation, and the abnormal values for these parameters are  $\geq 4.5$ , and  $> 2.5$  respectively(12). The abnormal value for AC is  $> 2.84$  (13).

**Statistical analysis**

Statistical analyses were performed using the Statistical Package for Social Sciences (SPSS Version 18.0). The demographic and continuous variables were presented as either mean  $\pm$  standard deviation or mean  $\pm$  standard error of the mean. To determine the differences between two means, the Student's t-test was utilized, while the Chi-square test was used to evaluate the degree of association between

dyslipidaemia and demographic factors, with Fischer's exact test applied when necessary. A P-value of less than 0.05 was considered statistically significant.

**Ultrasonography**

Any changes noted in serum lipids were correlated with ultrasonography of the liver. Accordingly, fatty liver is graded into Grade I fatty liver(GI FL), Grade II fatty liver (GII FL) and Grade III fatty liver(G III FL).Grade I fatty hepatomegaly(GI FH), Grade II fatty hepatomegaly(GII FH) and fatty hepatomegaly(FH) are the other categories related to fatty liver.

**RESULTS**

This study had a total of 150 subjects. The total number of males were 101 and females 49. Their ages ranged from 20-69 years with a mean age of  $45.53 \pm 11.81$ . The mean age of males was  $45.37 \pm 12.99$  and the mean age of females was  $45.87 \pm 8.99$ .

The distribution of dyslipidemia was analysed in the study population (Table 1). Of the study subjects, 22(15.0%) had low HDL-C. Interestingly all 22 were females. 63 (42%) had elevated total cholesterol (TC), 49 (33%) had elevated triglyceride levels, and 52 (35%) had elevated LDL cholesterol. In addition, 51 (34%) had elevated levels of non-HDL. Except for the observation of low HDL and elevated TG( $p=0.038$ ), there was no significant difference in other parameters between the sexes. 22.6%, 36.7%, 50.7% and 32.7% of the subjects had elevated TC/HDL, LDL/HDL, non-HDL/HDL and TG/HDL levels respectively

**Table 1:** Distribution of dyslipidemia among the study population

Lipid Profile/Lipid Ratios	Total (%)	Males (%)	Females(%)	P-value
Elevated TC $\geq 200$	63 (42%)	40 (27%)	23 (15%)	0.505
Elevated TG $\geq 150$	49 (33%)	39 (26%)	10 (7%)	0.038
Elevated LDL -C $\geq 130$	52 (35%)	30(20%)	22 (15%)	0.137
Elevated non-HDL -C $> 160$	51 (34%)	31 (21%)	20 (13%)	0.271
Elevated TC/HDL $\geq 4.5$	34(22.6%)	22 (14.6%)	12(8%)	0.710
Elevated LDL/HDL $> 2.5$	55 (36.7%)	32 (21.3%)	23 (15.3%)	0.074
Elevated TG/HDL $\geq 3.0$	49 (32.7%)	39 (26%)	10 (6.66%)	0.082
Low HDL $< 40$ men $< 50$ women	22(15.0%)	Nil	22(15.0%)	....

TC – Total cholesterol, TG – Triglyceride, LDL – C-Low-density lipoprotein cholesterol, Non-HDL-C – Non-HDL cholesterol, HDL-C- High-density lipoprotein –cholesterol.

The prevalence of dyslipidemia was observed to be higher between the ages of 30-49 years.

On analysing the lipid ratios 36.7% of subjects with an LDL/HDL ratio,  $> 2.5$  were in the age group 50-59 years and this was statistically significant ( $p=0.012$ ; Table 2).

**Table 2:** Age group and dyslipidemia

Age group	TC $\geq 200$	TG $> 150$	LDL $\geq 130$	N-HDL $> 160$	TC/HDL $\geq 4.5$	LDL/HDL $> 2.5$	Non-HDL/HDL $> 2.84$	TG/HDL $\geq 3.0$
20-29	1 (1.5%)	3(6.1%)	0 (0%)	1(1.9%)	0(0%)	0(0%)	1(1.3%)	3(6.0%)
30-39	15(23.8%)	16(32.6%)	8(15.3%)	10(19.6%)	5(14.7%)	8(14.5%)	18(23.6%)	16(32.0%)
40-49	20(31.7%)	13(26.5%)	19(36.5%)	17(33.3%)	10(29.4%)	19(34.5%)	26(34.2%)	12(24.0%)
50-59	19(30.1%)	11(22.4%)	18(34.6%)	16(31.3%)	13(38.2%)	20(36.3%)	22(28.9%)	13(26.0%)
60-69	7(11.1%)	5(10.2%)	6(11.5%)	6(11.7%)	5(14.7%)	7(12.7%)	8(10.5%)	5(10.0%)
$> 70$	1(1.5%)	1(2.0%)	1(1.9%)	1(1.9%)	1(2.9%)	1(1.8%)	1(1.3%)	1(2.0%)

<b>Total n (150)</b>	63 (42%)	49 (32.6%)	52 (34%)	51 (34%)	34 (22.6%)	55 (36.7%)	76 (50.7%)	50 (33.3%)
<b>Chi-squared p-value</b>	0.773	0.666	0.130	0.405	0.120	<b>0.012</b>	0.137	0.823

28% of the subjects had fatty liver of which 22% were males and 6% were females. Of the 42 subjects with

fatty liver, 12.6% had grade I FL, 4% had Grade II FL, 5.3% had GI FH and 4% had FH (Fig.1).

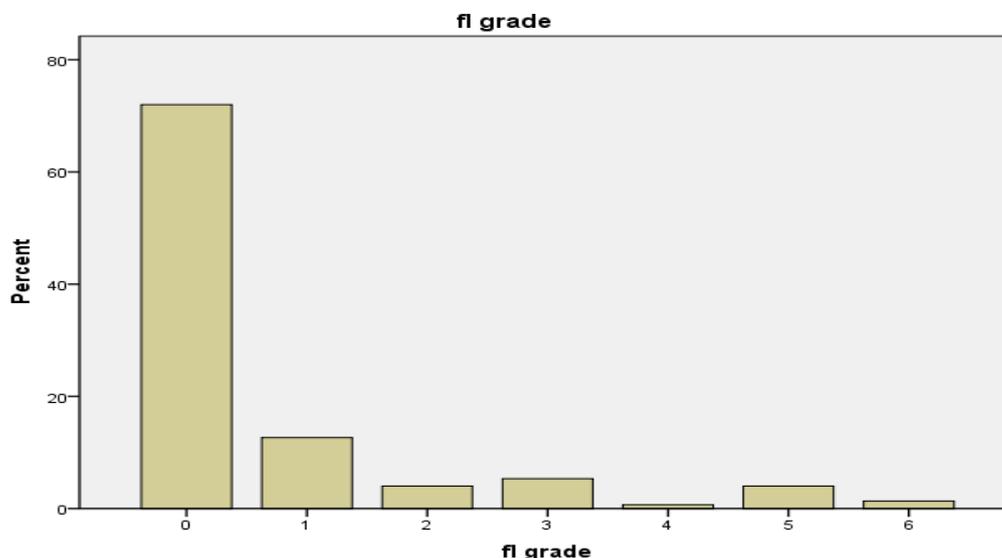


Fig 1. Distribution of different grades of fatty liver.

0 = no FL, 1- GI FL, 2- GII FL, 3- GI FH, 4-GII FH, 5-FH, 6-FL G-Grade, FL-Fatty liver, FH-Fatty Hepatomegaly

Across different fatty liver subtypes, 13/26(50%) of the subjects had elevated cholesterol levels and this was statistically significant (p=0.020). This was noted mainly in the subtype GI FL. The two ratios which

were significantly altered across the various subtypes were LDL/HDL (p=0.018) and non-HDL/HDL (p=0.000; Table 3).

Table 3: Fatty liver and prevalence of dyslipidemia

Fatty liver	Chol ≥200	TG ≥150	LDL ≥130	Non-HDL >160	TC/HDL ≥4.5	LDL/HDL >2.5	Non-HDL/HDL >2.84	TG/HDL ≥3.0
GI FL	13(50.0%)	6 (35.2%)	13 (65.0%)	10(47.6%)	8 (53.3%)	14 (62.6%)	18(52.9%)	7(36.8%)
GII FL	3 (11.5%)	3 (17.6%)	1 (5.0%)	2 (9.5%)	2(13.3%)	1 (4.5%)	5 (14.7%)	3 (15.7%)
GI FH	6 (23.0%)	4 (23.5%)	4 (20.0%)	4 (19.0%)	2(13.3%)	4 (18.1%)	6 (17.6%)	4 (21.0%)
GII FH	0	0	0	0	0	0	0	0
FH	3 (11.5%)	4 (23.5%)	2 (10.0%)	4 (19.0%)	3 (20.0%)	2(9.0%)	4(11.7%)	4 (21.0%)
FL	1 (3.8%)	1(5.8%)	0	1 (4.7%)	0	1 (4.5%)	1(2.9%)	1(5.2%)
Total n (%) 42 (28%)	26 (61.9%)	17 (40.4%)	20 (47.6%)	21(50.0%)	15(35.7%)	22 (52.3%)	34(80.9%)	19(45.2%)
<b>Chi squared p-value</b>	0.020	0.536	0.169	0.157	0.146	0.018	0.000	0.352

Across all the participants the mean of CRI (TC/HDL) was 3.925 ± 0.0603, CR II (LDL/HDL) 2.378 ± 0.0642 and AC (non-HDL/HDL) was 2.953 ± 0.0632. There was a significant difference in CR II between males and females (p=0.053). All the ratios were slightly

higher in females as compared to males. CRI, CR II, and AC indicated a cardiovascular risk prevalence of 22.6%, 36.7% and 50.7% respectively. There was no significant difference between the sexes (Table 4).

Table 4: Distribution of mean lipid ratios among the study population

Indices	Mean ± SEM			
	Total	Males	Females	P-value
CRI I TC/HDL	3.925±0.0603	3.905±0.0752	3.967 ± .1219	0.650
CRI II LDL/HDL	2.378±0.0642	2.297 ± 0.0720	2.545 ±0.1069	0.053
AC Non-HDL/HDL	2.953 ± 0.0632	2.930 ±0.0764	3.002 ± 0.1132	0.593

CR – Castelli’s Risk Index, AC- Atherogenic coefficient, SEM – Standard error mean

## DISCUSSION

Several studies have documented the prevalence of various forms of lipid disorders amongst Indians. Not only is the prevalence of dyslipidemia high amongst Indians but it has steadily increased in recent decades (14). Several studies conducted in India have reported a prevalence range of approximately 20% to 35% for hypercholesterolemia ( $TC \geq 200$  mg/dL) (15). In our study hypercholesterolemia was seen in 42% of the study group. Our findings were comparable to those of studies conducted in the rural populations of Central and Southern India, which reported a prevalence range of 37% to 57%. Our results were also similar to the global average of 39%, as reported by the World Health Organization (2). In addition, 32% had elevated TG, 34% had elevated LDL -C and 34% had elevated non-HDL. Low HDL was seen in 22 (15%) cases, and all were women. Low HDL amongst women was also observed in another recent study from Delhi (14). The percentage of elevated TG was higher in males than in females and this was statistically significant ( $p=0.038$ ).

The low HDL-C and hypertriglyceridemia are metabolically interlinked and their combination known as the 'atherogenic dyslipidemia' is particularly common amongst South Asians (15). The distribution of dyslipidemia was found to be highest between the ages of 40-49 yrs. Hypercholesterolemia (32%), elevated LDL (37%), elevated non-HDL (33%) and elevated non-HDL /HDL ratio (atherogenic coefficient) was the pattern of distribution in this group. Hypertriglyceridemia (33%) and increased TG /HDL ratio were mainly seen in the age group 30-39 years whereas LDL/HDL ratio (Castelli risk index II) was significantly elevated  $p=.012$  in the age group (50-59 years). Other studies also reported a higher age wise distribution of lipid abnormalities in the age group 51-60 years(16). Advancing age is usually listed as a major risk factor for dyslipidemia.

It is estimated that 25% of the population worldwide is affected by NAFLD (7). In our study, 28% of the subjects had fatty liver. Previous studies have shown that dyslipidemia and NAFLD are independently and strongly related (17) and patients with cardiovascular disease (CVD) risk factors including dyslipidemia are at high risk for NAFLD (18).

In our study, particularly in the Grade I fatty liver subtype 50% (13/26) of subjects had elevated cholesterol which was statistically significant ( $p=0.020$ ). 65% (13/20) had elevated LDL and 48% (10/21) had elevated non-HDL. The two ratios which were significantly elevated were LDL/HDL ( $p=0.018$ ) and non-HDL/HDL ( $p=0.000$ ). Our study corroborates that of Wang *et al.*, (19) that lipid ratios particularly non-HDL cholesterol to HDL cholesterol ratios could be an independent predictor of the onset of NAFLD.

The conventional strategy for managing coronary artery disease (CAD) risk centers on LDL-C, which is the primary atherogenic lipoprotein particle. However, LDL cholesterol has its limitations which makes it a questionable stand-alone marker for coronary artery disease (CAD) risk assessment (20). Non-HDL-C was introduced by ATP III guidelines in 2001, and several studies have strongly recommended its incorporation in the routine lipid profile panel (6). Instead of relying solely on the conventional lipid profile, Olamoyegun and colleagues (4) assessed dyslipidemia using lipid ratios and indices. In our study, the mean of CR II (LDL/HDL) and AC (non-HDL/HDL) were higher than their normal cut off values. There was a significant difference in CR II (LDL/HDL) between males and females. ( $p=0.053$ ). All the lipid ratios were slightly higher in females than in males. This has also been observed in another study(4). In our study group, the most prevalent form of the atherogenic index was the Atherogenic coefficient (non-HDL/HDL) identified in 50.7% of the population. The prevalence rate of AC is higher in our subjects compared to the results of another study conducted in Nigeria. (4). Studies have shown that non-HDL is analogous to Apo -B and both non-HDL-C and Apo -B has a high correlation with CAD risk. The atherogenic non-HDL-C fraction is composed of chylomicrons, very-low-density lipoproteins (VLDL), intermediate-density lipoproteins (IDL), and low-density lipoproteins (LDL) (20). Without a doubt, the atherogenic coefficient would reflect the atherogenic potential of the entire spectrum of lipoprotein fractions and therefore the cardiovascular risk (21).

## CONCLUSION

Our study highlights that even in rural populations dyslipidemia is a matter of concern especially as the subjects had come for a routine check-up with no prior history of any chronic disease. The present study clearly indicates that lipid ratios play a very important role in early screening and should be incorporated into the screening protocol. This may play an important role in reducing the risk of future coronary artery disease and fatty liver disease.

## CONFLICT OF INTEREST

Authors declare no conflicts of interest for this study.

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