

Calculated lipid parameters, flip side to determine hypercholesterolemia

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ABSTRACT

Objective: To determine that calculated lipid parameters have more diagnostic value in Hypercholesterolemia.

Material and Methods: This comparative cross-sectional study conducted from October 2023 till April 2024 in tertiary care hospital Kharian. Total 414 individuals were included in this study who reported for Lipid Profile. Samples for fasting lipid profile including serum cholesterol, Triglycerides, HDL-cholesterol and LDL-cholesterol, were collected and analyzed by automated chemistry analyzer. Individuals with Total Cholesterol were sub categorized into 2 groups (Group I < 5.2 mmol/L and group II > 5.2 mmol/L). However, lipid parameters were estimated by different calculated formulas.

Results: Out of 414 participants of the study, 296 (71.5%) had cholesterol level < 5.2 mmol/L and 118 (28.5%) presented with cholesterol level > 5.2 mmol/L. 330 (74.8 %) were male and 84 (25.2%) were female. Mean age with SD was 44.48 ± 12.78 years. There was statistically significant high association (p value < 0.005) between calculated non-HDL and Hypercholesterolemia as compared to other calculated and measured parameters.

Conclusion: Calculated non- HDL parameter is clinically more valuable markers to diagnose hypercholesterolemia in addition to good prognostic and treatment goal.

Keywords: Cholesterol ratios, Hypercholesterolemia, LDL-cholesterol, non-HDL cholesterol, Triglycerides.

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INTRODUCTION

Hypercholesterolemia, raised cholesterol level in the blood, is a substantial threat for heart diseases, including heart attacks and strokes [1]. Conventionally, high-density lipoprotein (HDL) cholesterol has been acclaimed as the "good cholesterol," while low-density lipoprotein (LDL) cholesterol has been maligned as the "bad cholesterol [2]." Approximately 18-25% of adults in Pakistan have raised cholesterol, particularly in urban areas. In Asia, countries like China and India, hypercholesterolemia is projected to be around 20-30% among adults [3]. According to the World Health Organization (WHO), about 39% of adults aged 25 years and older had raised total cholesterol levels globally. This interprets to over 1 billion adults worldwide. Moreover, hypercholesterolemia is accountable for a substantial burden of morbidity and mortality, contributing to millions of deaths annually [4].

Cholesterol is transported in the bloodstream by lipoproteins, with HDL and LDL. HDL cholesterol is deliberated helpful because it conveyances cholesterol from the arteries to the liver for elimination, hence decreasing the threat of plaque formation. Contrariwise, LDL cholesterol is known for dropping cholesterol in the arterial walls, leading to atherosclerosis and increased risk of CVDs [5]. Non-HDL cholesterol includes all cholesterol except HDL, like apolipoprotein B (apo B), very low-density lipoproteins (VLDL) and their metabolic leftovers, intermediate density lipoproteins (IDL), lipoprotein(a), and low-density lipoproteins LDL cholesterol.

Non-HDL-C can be called comprehensive atherogenic indicator as it includes all atherogenic lipoproteins. It provides comprehensive assessment of cardiovascular risks in all condition of dyslipidemia particularly in metabolic syndrome and type II diabetes. American Heart Association and the National Lipid Association accentuate its role in primary and secondary prevention. In primary and secondary prevention cut off of non-HDL-C are proposed to be 3.36 mmol/L and 2.59 mmol/L respectively with reference interval of 2.59-4.91 mmol/L [6].

Non-HDL-Cholesterol = Total Cholesterol - HDL-Cholesterol

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Friedewald formula was introduced by William Friedewald in 1972. Despite its limitation like non-fasting sample and high triglycerides level, it is most convenient and cost effective in determining LDL-C. Due to its limitation, non-HDL-C is making its importance more. In primary and secondary prevention cut off of LDL-C are suggested to be 3.36 mmol/L and 2.59 mmol/L respectively [7].

LDL-Cholesterol = Total Cholesterol – HDL-Cholesterol - Triglyceride/ 2.21 **(Friedewald formula) (Triglyceride level < 4.5 mmol/L)**

Modified Friedewald formula was introduced to calculate LDL-C by introducing HDL-C and correction factors in formula to overcome the limitations of Friedewald formula and allowing for more accurate risk stratification and management of cardiovascular disease [8].

LDL-Cholesterol = Total Cholesterol/1.19 + Triglyceride/0.81- HDL-Cholesterol/1.1- 0.98 **(Modified Friedewald formula)**

de Cordova formula was introduced by Dr. C. M. de Cordova to improve the accuracy of calculated LDL-C in population other than in which Friedewald formula originally designed. LDL-C calculated by de Cordova formula is more align to measured LDL-C than LDL-C calculated by Friedewald formula in patients with high triglycerides level 7 [9].

LDL-Cholesterol = 0.75 (Total Cholesterol- HDL-Cholesterol) **(de Cordova formula)**

Calculated lipid ratios offer an abridged methodology to interpret lipid profiles by providing a holistic view of atherogenic and protective lipoproteins. These ratios also serve as important clinical tools for risk stratification and therapy decision-making in hypercholesterolemia. In primary and secondary prevention cut off of Cholesterol ratio are suggested to be 5 and 4 respectively [10].

Cholesterol Ratio = Total Cholesterol ÷ HDL-Cholesterol **(Castelli risk index I)**

Cholesterol Ratio = LDL-Cholesterol ÷ HDL-Cholesterol **(Castelli risk index II)**

Therefore, in this study we aimed to compare Non-HDL-C (both measured and calculated) and other lipid calculator parameters in patients with high Cholesterol level to determine which parameter is more valuable by a cross-sectional study. Furthermore, it is a cheaper option

MATERIAL AND METHODS

A Comparative Cross-sectional study conducted in October 2023 to April 2024, at the Chemical Pathology and Endocrinology department, tertiary care hospital, Kharian after permission from the review board of institution vide letter number: A/24/22 dated 10th October 2023. Non-probability consecutive technique was employed (considering 32.2% [11] prevalence of hypercholesterolemia with 7% margin of error and 95% of confidence interval) and sample size was found to be 414 as calculated by Open Epi version. Study participants were chosen based on inclusion and exclusion criteria. Inclusion criteria comprised individuals aged 18 years and older having family history of hypertension, cardiac disease, stroke, drugs and prone to develop all these diseases themselves were included. All those who were on lipid lowering drugs were excluded from our study.

After taking relevant history, procedure was explained to all individuals. Sample for fasting lipid profile was collected in gel-clot activator tube. Specimen was allowed to clot for 30 minutes and centrifugation was performed. Samples were analyzed by automated chemistry analyzer based on photometry for all lipid profile parameters including serum total cholesterol, serum triglyceride, serum LDL-Chol, serum HDL-Chol. Non-HDL cholesterol and LDL-cholesterol were calculated by using formulae mention above. Data was collected using a dedicated structural proforma from study participants after getting informed consent. Clinical and demographic surveys was conducted to collect important information regarding age, gender, smoking and past medical history and all information was noted down.

Data was analyzed by SPSS 21. The normality of the data was assessed using the Kolmogorov Smirnov test. Blood pressure (BP), Total Cholesterol, LDL and HDL were found to be normally distributed ($p > 0.05$), whereas age, BMI, and fasting glucose were non-normally distributed ($p < 0.05$). Continuous variables with a normal distribution (e.g. BP, LDL, and HDL) were presented as mean (standard deviation) (SD), while non-normally distributed variables (e.g., age, BMI, fasting and glucose) were presented as median with interquartile range (IQR). Qualitative variables (age, group, BMI categories, gender, smoking status, Hypertension, diabetes mellitus, previous stroke history) were presented as frequency and percentages. Descriptive statistics were calculated from study variables. Quantitative variables like

age, weight, height, BMI, duration were presented as Mean and Standard Deviation. Percentage and frequency were calculated for qualitative variables. Spearman test was applied to compare mean and Chi-square test were applied to association between non-HDL cholesterol and hypercholesterolemia. ANOVA was also applied to observe relationship of LDL cholesterol calculated by different formulae and with non-HDL cholesterol. taking p-value < 0.05 as statistically significant.

RESULTS

Four hundred and fourteen individuals of different ages were included in the study. Mean age with SD was 44.47 ± 12.77 years. 330 (74.8 %) were male and 84 (25.2%) were female. Out of 414 participants, 186 (42.2%) were belonged to 41 – 60 years the age group. In all participants of study, 296 (71.5%) had cholesterol level < 5.2 mmol/L and 118 (28.5%) presented with cholesterol level > 5.2 mmol/L.

Median with IQR value of serum total Cholesterol, Triglyceride, LDL-Chol, HDL-Chol and calculated parameters including LDL-cholesterol and non-HDL-Chol were 4.6 (1.5) mmol/L, 1.7 (1.4) mmol/L, 2.8 (1.4) mmol/L, 1.0 (0.3) mmol/L, 2.3 (1.4) mmol/L, 4.1 (1.8) mmol/L, 2.6 (1.3) mmol/L, 4.4 (1.9) mmol/L, 2.6 (1.4) mmol/L and 3.5 (1.7) mmol/L respectively (as shown in Table-II).

One sample Kolmogorov-Smirnov test between calculated Total cholesterol and non-HDL-Chol was used with p value < 0.05, indicated data was non-parametric. Mean age of participants are 42 years and 43 years with cholesterol level of < 5.2 and > 5.2 mmol/L. Correlation coefficient of calculated non-HDL cholesterol and LDL-cholesterol with total cholesterol is 0.536 and 0.822 indicating statistical significance of non-HDL parameter. Beta value of -41.87 represent standardized coefficient. -1.000 indicating partial correlation relationship between total cholesterol and non-HDL cholesterol. 2.442E-005 indicating very high degree of collinearity statistics tolerance (as shown in Table-III). Spearman correlation between total cholesterol and different calculated parameters as per age and gender indicated statistical significance (as shown in Table-I). One sample ANOVA among total cholesterol different calculated parameters also indicated statistical significance level. Multiple linear regression keeping total cholesterol as a dependent and non-HDL cholesterol as independent variable indicated p value < 0.05 statistically significant (as shown in Figure-I, II & III). Multiple Linear regression having standardized and unstandardized -0.015 and -0.017 with tolerance -0.734 indicating clinically value able. (as shown by Table-III).

Table-I: Distribution of lipid profile parameters as per total cholesterol level.

Parameters	Mean ± SD	Serum Cholesterol level mmol/L		p Value
		< 5.2	>5.2	
Age (Years)	44.48 ± 12.78	43	44	0.000
Total Cholesterol mmol/L	4.69 ± 1.43	-	-	0.000
Triglycerides mmol/L	2.47 ± 0.27	1.6	2.6	0.000
HDL-Cholesterol mmol/L	1.09 ± 0.40	1.0	1.1	0.000
LDL-Cholesterol (measured) mmol/L	2.99 ± 1.44	2.5	4.1	0.000

Table-II: Distribution of calculated cholesterol parameters as per total cholesterol level.

Parameters	Median ± IQR	Correlation coefficient R	Serum Cholesterol Level mmol/L		p Value
			< 5.2	>5.2	
Non-HDL-Cholesterol mmol/L	3.5 ± 1.7	0.536	2.9	4.8	0.002
LDL-Cholesterol (Calculated FW) mmol/L	2.3 ± 1.4	0.716	2.1	3.4	0.000
LDL-Cholesterol mmol/L (Calculated modified FW)	4.1 ± 1.8	0.822	3.4	5.7	0.000
LDL-Cholesterol mmol/L (Calculated de Cordova)	2.6 ± 1.3	0.923	2.2	3.6	0.002
Castelli risk index I	4.4 ± 1.9	0.622	3.8	5.7	0.000
Castelli risk index II	2.6 ± 1.4	0.623	2.6	3.5	0.000

Table-III: Correlation among different calculated lipid parameters.

Parameters	Unstandardized Coefficients		Standardized Coefficients	Tolerance T	p value
	B	Std Error	Beta		
Non-HDL-Chol	-0.015	0.029	-0.017	-0.734	0.003
Calc LDL-Chol	-0.014	0.036	-0.012	-0.397	0.692
LDL Chol FW	0.006	0.032	0.006	0.201	0.841
LDL de Cordova	1.661	0.059	1.269	28.023	0.000

LDL Chol Mod FW	-	-	- 41.872	-2316610	0.000
Castelli risk index I	- 0.186	0.028	-0.283	- 6.571	0.000
Castelli risk index II	- 0.095	0.036	- 0.095	- 2.619	0.009

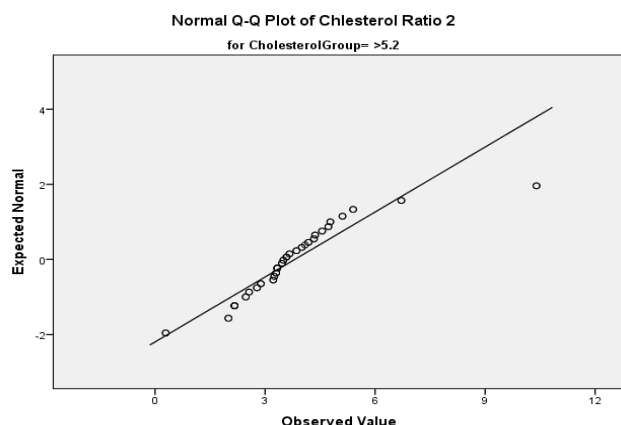


Figure-I: Line diagram indicating correlation of hypercholesterolemia and cardiovascular events

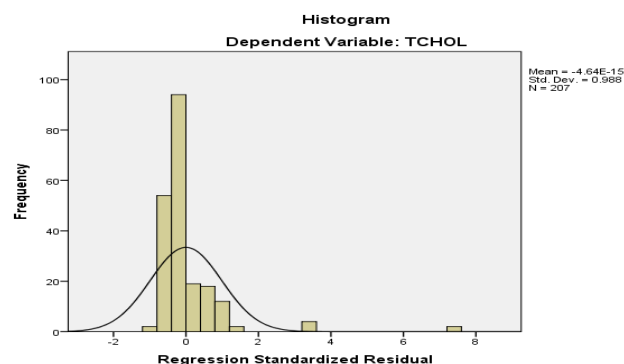


Figure-II: Histogram indicating correlation of hypercholesterolemia and cardiovascular events as per age

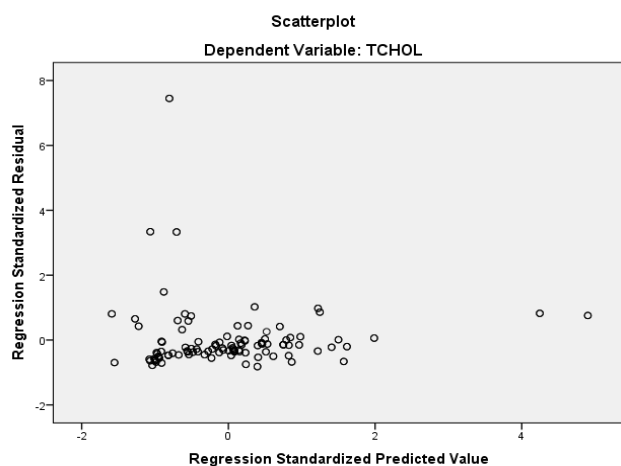


Figure III: Scatter diagram indicating correlation of hypercholesterolemia and cardiovascular events as per gender

DISCUSSION

In our study clinical value of calculated lipid parameters were determined. 414 patients of different ages were included in our study in which 330 (74.8 %) were male and 84 (25.2%) were

female. Out of 414 participants, 186 (42.2%) were belonged to of 41 – 60 years the age group. Fasting lipid profile including serum cholesterol, Triglycerides, LDL-C and HDL-C were analyzed. Calculated parameters were determined by using lipid measured parameters by different formulae. Correlation among these calculated lipid parameters to diagnose hypercholesteremia was determined. Correlation coefficient of calculated non-HDL cholesterol and LDL-cholesterol with total cholesterol is 0.536 and 0.822 indicating clinical value of non-HDL parameter All these calculated parameters showed positive correlation to diagnose hypercholesteremia (as shown in Table II). These parameters are also compared to each other and calculated non-HDL parameter is the most clinical valuable among all calculated parameters in diagnosing hypercholesteremia which can be supported and enforced statistical by p value (as shown in Table II). Diagnostic value for hypercholesteremia further increased by combining these parameters.

In our study we found that raised value of non-HDL are related with a bigger risk of mortality, representing that this marker is an easy and dependable tool for evaluating antagonistic consequences in the general population as well as in cardiovascular patients. Additionally, patients with increased non-HDL levels had a greater cardiovascular risk autonomously of their LDL-Cholesterol levels [12], and non-HDL was recognized superior in its predictive value when compared to LDL-Cholesterol, interestingly, the EPIC [13] (European Prospective Investigation into Cancer and Nutrition) study stated that even in the group of individuals with LDL-Cholesterol below cut off value [14,15].

The Castelli indices, compared with non-HDL-Cholesterol, have high correlation between is possibly because most of the cholesterol is present in LDL-Cholesterol [16], thus Total Cholesterol and LDL-Cholesterol are closely associated. Similarly, variations in both indices are revealed to be better indicators for reduction of cardiovascular risk when compared to other discrete lipids parameters [17] which is indicated with correlation value of our study. LDL measured by Friedewald, modified Friedewald and de Cordova formulas have some limitations like non-fasting sample, TG > 4.5 mmol/L < 0.8 mmol/L and co-morbidities like Diabetes, kidney and liver

diseases. In a study [18] clinical predictive value of LDL-C and non-HDL-C was compared. In this study effect of statins on level of non-HDL and LDL was observed. Non-HDL-C was markedly increased relative to LDL-C in potentially risked patients, recommending that more decrease in non-HDL-C than LDL-C would be needed for optimal risk factors management [19, 20]. The finding of study mentioned above gave supporting evidence to use non-HDL cholesterol as target of therapy. As per correlation coefficient value determined in our study, non-HDL cholesterol is better marker than LDL-cholesterol [21], similarly it is mentioned in literature [22], that Non-HDL-C foreseen cardiovascular mortality and morbidity better than LDL-C. With high levels of non-HDL-C conforming to a high risk of cardiovascular mortality and morbidity [23]. Furthermore, non-HDL-C pointedly foreseen cardiovascular mortality and morbidity. Actually, LDL-C levels were the minimum dependable predictor of cardiovascular mortality and morbidity, when compared with non-HDL-C [24].

CONCLUSION

Non-HDL-C is documented as an appreciated screening marker in hypercholesterolemia because of its straightforwardness, acceptance, impartiality, objective, not requirement of fasting state, and greater prognostic value. It measures accurate risk of atherogenicity than any other lipoprotein constituent.

CONFLICT OF INTEREST

Authors declare no conflict of interest.

GRANT SUPPORT & FINANCIAL DISCLOSURE

Declared none

AUTHORS CONTRIBUTION

Waqas Hanif: Study design, acquisition of data, Manuscript drafting or reviewing it critical for important intellectual Content, final approval, accountable for all aspects of publication.

Muhammad Shahzad Anwer: Manuscript drafting or reviewing it critical for important intellectual Content final approval, accountable for all aspects of publication.

Javaria Ahsan: Substantial contributions to analysis and interpretation of data, Critical review

Muhammad Aurangzeb: Manuscript drafting or reviewing it critical for important intellectual Content,

final approval, accountable for all aspects of publication.

Syed Mohsin Manzoor: Substantial contributions to concept, study design, final approval, accountable for all aspects of publication.

Urwa Sarwar: Analysis and interpretation of data, final approval, accountable for all aspects of publication

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