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Original Article

Biochemistry Section

Comparison of LDL Levels Obtained by Martin-Hopkins Formula and Friedewald's Formula with Directly Measured LDL: A Cross-sectional Study

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ABSTRACT

Introduction: Low Density Lipoprotein (LDL) is one of the major modifiable risk factor of atherosclerotic diseases and so a potential therapeutic target. So, Low Density Lipoprotein Cholesterol (LDL-C) is of primary importance to assess cardiac risk. Many formulae are used to calculate LDL-C. Friedewald's formula, despite its limitations, has been widely used due to its simplicity and convenience. The present research was aimed to compare the accuracy of Friedewald's formula with the Martin-Hopkins formula, providing valuable insights for clinical practice.

Aim: To compare LDL levels obtained by the Martin-Hopkins and Friedewald's formula with LDL levels obtained by directly measuring LDL in the southern India.

Materials and Methods: The present hospital-based cross-sectional study was conducted in the Department of Biochemistry, Hassan Institute of Medical Sciences (HIMS), Hassan, Karnataka, India, from March 2023 to June 2023. Total participants were assessed for their serum lipid profile using standard methods. LDL-C was calculated using the Friedewald and Martin formulae, and these calculated values were then correlated with the directly measured LDL-C. Based on Triglyceride (TG) levels, subjects were divided into five groups to ensure a comprehensive analysis (Group-1: TG <100 mg/dL, Group-2: TG=101-150 mg/dL, Group-3: TG=151-200 mg/dL, Group-4: TG=201-400 mg/dL, Group-5: TG >400 mg/dL). Groups were compared using a correlation analysis using Pearson's correlation and paired t-test.

Results: The study findings are significant. At TG levels <100 mg/dL, the Martin formula demonstrated a superior correlation (r=0.964, p-value ≤0.001). At TG 101-150 mg/dL, 151-200 mg/dL, and 201-400 mg/dL, Friedewald's formula showed a slightly better correlation. However, at TG >400 mg/dL, Friedewald's formula had a higher mean difference. ROC curves further confirmed the superiority of Martin's formula (AUC=0.948, p-value ≤0.001), demonstrating better diagnostic performance than Friedewald's formula (AUC=0.947, p-value ≤0.001) at all TG levels.

Conclusion: The Martin-Hopkins formula showed better diagnostic performance among the two equations than the Friedewald's formula. The mean difference was lesser for Martin's formula than Friedewald's formula at all TG levels except at levels <100 mg/dL. Considering the mean difference, Martin's formula provided better LDL-C values than Friedewald's formula for estimating LDL-C in the present study's demographic population.

Keywords: Cardiovascular disease, Lipid profile, Low density lipoprotein, Triglyceride

INTRODUCTION

Cardiovascular diseases account for major share of global mortality [1]. Among markers of cardiovascular risk assessment, lipoprotein levels have played a vital role for a long-time [2,3]. Cholesterol is carried from the liver to peripheral tissues by LDL-C. LDL-C has proatherogenic properties, making it a major modifiable risk factor and therapeutic target for atherosclerotic disease. It is clinically calculated using Friedewald's formula worldwide [4]. β-quantification and ultracentrifugation is the accepted gold-standard method for LDL measurement although labour-intensive, time-consuming, and expensive [5].

Lipid profile tests consist of measurement of TG, High-density Lipoprotein Cholesterol (HDL-C), Total Cholesterol (TC) and calculated LDL (LDLcal). Friedewald equation is used to calculate LDL {LDL Friedewald=TC-HDL-(TG/5)} [6]. However, the Friedewald equation is inaccurate at TG concentrations 200-400 mg/dL and becomes invalid at TG levels >400 mg/dL [7]. So, based on studies, several LDL calculation equations have been suggested as alternatives to Friedewald [8,9].

Martin SS et al., proposed a new equation for LDL estimation (LDLMartin) using an adjustable factor for TG: Very Low-Density Lipoprotein Cholesterol (VLDL) ratio based on TG levels and concentration of non HDL (non HDL=TC-HDL) [4]. They observed that the overall concordance in risk classification with LDL direct for patients with TG lower than 400 mg/dL was 85.4% for LDL Friedewald and 91.7% for LDL Martin [8].

In clinical conditions such as chronic renal failure on dialysis, alcoholic liver disease, and diabetes mellitus Friedewald's equation either underestimates or overestimates LDL-C [10,11]. Over or under estimation of LDL-C causes problems to patients. Overestimation leads to prescription of unnecessary medication, underestimation delays the necessary therapy, leading to an increased cardiac threat [10,12]. So, many attempts were made by researchers to modify the equation with varied success [10,12]. This variation is possible due to the differences in the populations used to derive the equation, which vary in ethnicity, environmental influences, and demographics. However, each equation provides a different result. This suggests a need to expand the availability of alternative, more accurate and reliable techniques or methods that can be used to calculate LDL-C, when direct LDL-C estimation equipment and reagent kits are not available.

Based on the vertical auto profile test, an ultracentrifugation-based method, an equation was developed by Martin SS et al., which will be stated hereafter as Martin equation [8]. The Martin equation showed better accuracy for low-LDL-C samples than the Friedewald equation [13]. It uses an adjustable factor based

on the concentration of non HDL-C and TG, instead of a fixed TG denominator of five in the Friedewald equation. Limited data on the accuracy of equations for LDLcal are available in the Indian population [10,14]. LDL Friedewald is most commonly used for LDL calculation. The current study was aimed to verify the performance of Martin-Hopkins and Friedewald formulae with directly measured LDL among South Indians to improve the utility of calculated LDL when direct LDL measurement is not available.

MATERIALS AND METHODS

A hospital-based cross-sectional study was carried out in the Central Laboratory, Department of Biochemistry, HIMS teaching hospital, Hassan, Karnataka, India, from March 2023 to June 2023. Institutional Ethics Committee clearance was obtained (ethical approval Ref No IEC/HIMS/RR 391/07/03/23). Objectives and procedure of study were explained to all study participants and informed consent was obtained.

Sample size calculation: As per a previous study, the prevalence of lipid abnormalities was 79%, with a sensitivity and specificity of LDL/HDL ratio was 65% and 61%, respectively [15,16].

$$Se=\frac{Z^2_{\alpha/2}Se\ (1\text{-}Se)}{d^2\times prev}\quad nSp=\frac{Z^2_{\alpha/2}Sp\ (1\text{-}Sp)}{d^2\times(1\text{-}prev)}$$

For α=0.05, the value of Z for α/2 was taken as 1.96. Sensitivity (Se), specificity (Sp), and prevalence (prev) were the predetermined values, and d represented the precision of the estimate (i.e., the maximum marginal error) determined by clinical management of investigations. Therefore, the sample size for the present study was 400.

Inclusion criteria: All subjects over 18 years who visited hospital laboratory for fasting lipid profile test were included in the study.

Exclusion criteria: Patients with diabetes mellitus, thyroid dysfunction, pregnant women, chronic kidney disease, chronic hepatitis, cirrhosis, pancreatitis and patients on steroids, omega-3 fatty acids and statins medication, as well as, paediatric cases, were excluded from the study.

Study Procedure

Fasting samples for lipid profile test were collected from 400 study participants. The lipid profile of the participants was categorised, and HDL-C, TGs, LDL-C, and TC levels were estimated in the provided samples. Additionally, they were divided into five groups based on TG levels (Group-1: TG <100 mg/dL, Group-2: TG=101-150 mg/dL, Group-3: TG=151-200 mg/dL and Group-4: TG=201-400 mg/dL, Group-5: TG >400 mg/dL) [10].

Study tools and study variable: Demographic information, personal history, statin therapy details were recorded from the participants by using a structured case report form. The study variables included lipid profile parameters such as TG, LDL-C, TC and HDL-C were included as the study variables.

Lipid measurements: Fasting blood samples were collected after an overnight fasting period of 10-12 hours, 3 mL of venous blood was drawn into a plain tube. The samples were then centrifuged at 3200 rpm for 10 minutes for separation of serum and analysed immediately using the Abbott Architect ci4100 integrated chemistry and immunoassay analyser. The serum lipid profile parameters included TG, HDL-C, TC and LDL-C. All lipid parameters were measured using kits purchased from Abbott Architect system packs to determine direct LDL-C (measured using a Liquid selective detergent), HDL-C (accelerator selective detergent), TG (glycerol phosphate oxidase) and TC (enzymatic method).

Standard quality control practices as per ISO 15189 were in place to ensure reliability of results. Liquichek Lipids Control internal quality controls from Randox laboratories, Inc were assayed for monitoring of precision. Laboratory participated in Randox International Quality Assessment Scheme (RIQAS) external quality assurance programme conducted by Randox laboratories to ensure the accuracy of the analytical testing.

Methods used to calculate LDL-C:

- Martin-Hopkins formula: LDL=TC-HDL-TG/novel factor derived using an LDL-C calculator [8]. LDL-C calculator from http://www.ldlcalculator.com was used to calculate Martin's LDL-C. The TC, HDL-C and TG values were entered into the Microsoft Excel file for calculation of LDL-C and non HDL-C, and the adjustable factor by Martin's formula was automatically calculated.

TG and non HDL-C concentrations decide the adjustable factor. To estimate VLDL-C cholesterol from TGs, Martin's employs a technique that matches each person with 1 of 180 factors.

- Friedewald formula: LDL-C=TC-HDL-C-TG/5 [17].

STATISTICAL ANALYSIS

Statistical analysis was performed using Epi Info software version 7.2. The data obtained were entered into a Microsoft Excel sheet. The data were expressed as mean and standard deviation. The lipid profile parameters were correlated by use of Pearson's correlation test. A paired t-test was performed to compare the means of biochemical parameters between the groups. The diagnostic performance of Martin's and Friedewald's formulas was evaluated by use of Area Under Curve (AUC), which was obtained by use of Receiver Operating Characteristic (ROC) Curves. A two-tailed p-value of <0.05 was considered statistically significant.

RESULTS

A study of 400 participants was conducted, including 246 females and 154 males. The population was divided into five groups based on TG values. The mean TC was 182.29±42.220 mg/dL, while LDL-C levels were 128.76±38.856 [Table/Fig-1]. The calculated formulas underestimated LDL-C by 6 mg/dL and 23 mg/dL using Martin's and Friedewald's methods, respectively, compared to the direct method.

Variables	Mean±Standard deviation
Mean age (years) of total number of subjects included in the study	41.16±12
Males (Age in years)	39.40±12.3
Males (Age in years)	43.97±12.5
Serum TC (mg/dL)	182.29±42.220
Serum HDL-C (mg/dL)	39.319±0.479
Serum triglycerides (mg/dL)	187.60±118.850
Serum direct LDL-C (mg/dL)	128.76±38.856
Serum Martin-Hopkins (M-LDL-C, mg/dL)	112.6±35.029
Serum Friedewald's (F-LDL-C, mg/dL)	105.46±36.89

[Table/Fig-1]: Demographic distribution and data of lipid profile of study subjects.

Direct LDL-C with calculated LDL-C using Friedewald's and Martin's formulas are compared in [Table/Fig-2]. Both formulas consistently underestimate LDL at all TG levels, with a statistically significant difference (p-value <0.001). Notably, Martin's formula showed a smaller mean difference between direct and calculated formulas across the total sample, except for group-1.

Group	Variable	Mean±SD	Mean difference (between direct and calculated LDL)	t-test p-value
Group-1	LDL	117.23±33.05	-	-
	Martin's	102.65±26.64	14.58±10.26	<0.001
	Friedewald's	104.54±26.96	12.68±10.19	<0.001
Group-2	LDL	132.23±30.32	-	-
	Martin's	111.06±28.18	21.17±17.24	<0.001
	Friedewald's	110.62±26.82	21.60±9.85	<0.001

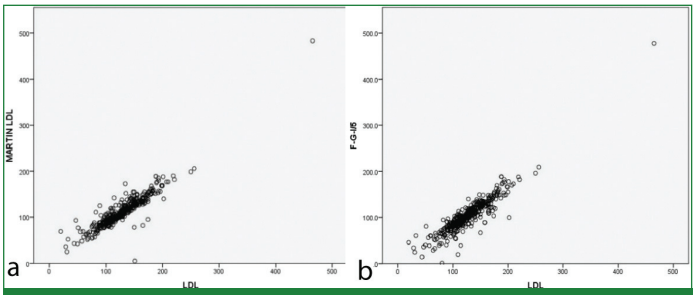
Group-3	LDL	126.64±35.04	-	-
	Martin's	112.47±32.33	14.16±10.55	<0.001
	Friedewald's	106.19±34.47	20.44±10.14	<0.001
Group-4	LDL	135.32±48.77	-	-
	Martin's	119.74±45.15	15.58±16.22	<0.001
	Friedewald's	106.41±48.28	28.90±14.19	<0.001
Group-5	LDL	127.70±41.82	-	-
	Martin's	119.65±31.30	8.05±28.40	0.220
	Friedewald's	74.74 ±36.43	52.96±25.29	<0.001
Total sample	LDL-C	128.76±38.85	-	-
	Martin	112.68±35.02	16.2±3.5	<0.001
	Friedewald's	105.46±36.89	23±2.5	<0.001

[Table/Fig-2]: Comparison of mean value of direct LDL-C and calculated LDL-C by Martin and Friedewald's formulae.
SD: Standard deviation; LDL-C; Low-density lipoprotein cholesterol; Mean difference=Direct LDL cholesterol- formula-calculated LDL cholesterol; The p-value <0.05 was considered statistically significant

The LDL-C by Martin formula showed a better correlation with direct LDL (r-value=0.964, p-value <0.001) compared with Friedewalds (r-value=0.963, p-value <0.001) in group-1. LDL-C by Friedewald formula showed a slightly better correlation with direct LDL than Martin formula in all other groups [Table/Fig-3,4].

Samples	Formula	r-value	p-value
Group-1	Martin's formula	0.964	<0.001
	Friedewald's	0.963	<0.001
Group-2	Martin's formula	0.829	<0.001
	Friedewald's	0.948	<0.001
Group-3	Martin's formula	0.954	<0.001
	Friedewald's	0.958	<0.001
Group-4	Martin's formula	0.943	<0.001
	Friedewald's	0.957	<0.001
Group-5	Martin's formula	0.734	<0.001
	Friedewald's	0.800	<0.001
Total	Martin's formula	0.916	<0.001
	Friedewald's	0.920	<0.001

[Table/Fig-3]: Correlation between direct LDL-C and calculated LDL-C by Martin and Friedewald's formulae and Karl Pearson's correlation method.
The p-value <0.05 was considered statistically significant

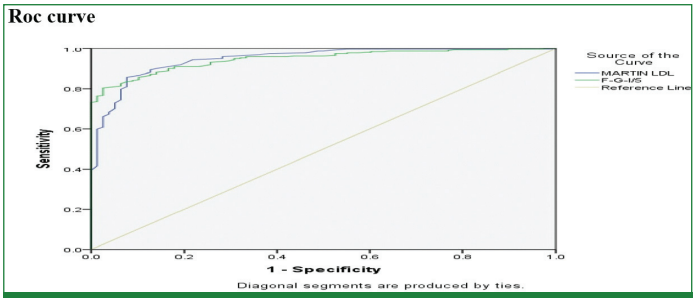


[Table/Fig-4]: Correlation between direct Low-Density Lipoprotein-cholesterol (LDL-C) and calculated LDL-C. a) Correlation between direct low-density lipoprotein-cholesterol LDL-C and Martin formula LDL-C. b) Correlation between direct Low-Density Lipoprotein-Cholesterol (LDL-C) and Friedewald's formula LDL-C.

Diagnostic performance: To analyse the performance of calculated LDL-C using Martin's and Friedewald's formulas ROC curves were constructed [Table/Fig-5,6]. Out of the two formulas, Martin-Hopkins (AUC=0.948, p-value <0.001) exhibited better diagnostic performance, followed by Friedewald (AUC=0.947, p-value <0.001).

Calculated LDL-C	AUC	p-value
Martin LDL	0.948	<0.001
Friedewald LDL	0.947	<0.001

[Table/Fig-5]: Area Under Curves (AUC) of calculated LDL-C using the two formulae.
AUC: Area under curve; The p-value <0.05 was considered statistically significant



[Table/Fig-6]: Diagnostic performance of two formulae. ROC: Receiver operating characteristic.
Martin's Formula: Sensitivity (90%), Specificity (86%); Friedewald's Formula: Sensitivity (90%), Specificity (83%)

DISCUSSION

The present study found that both the Friedewald and Martin formulas consistently underestimated LDL-C across various TG groups. The present study also revealed that Friedewald's equation showed a higher mean difference with D-LDL in groups-2, 3, and 4 when all groups were considered simultaneously. This is a significant finding as it adds to the growing evidence that Friedewald's equation's performance diminishes with increasing TG levels. This finding is in line with previous research by Gupta S et al., and Agrawal M et al., [17,18].

The previous studies by Miller WG et al., and Nakanishi N et al., have shown that the mean difference between direct and formula-calculated LDL-C increases as TG levels increase [7,19]. The present study results support this finding: with increased TG concentrations, the difference between Direct LDL-C (D-LDL-C) and LDL-C calculated by Friedewald's and Martin's formulas increased. The current study recorded similar results. Other studies by Gupta S et al., Vujovic A et al., and Lindsey CC et al., which measured the LDL-C using different homogenous assays align with this [17,20,21].

Guidelines from European Society of Cardiology and European Atherosclerosis Society, American Heart Association and American College of Cardiology, Canadian Cardiovascular Society, the Heart, Lung, and Blood Institute, and National Cholesterol Education Programme (NCEP) guidelines assign the highest level of evidence (class 1A) to the LDL-C treatment goal [22-28]. So, more accurate measurement of LDL-C is essential to prevent adverse patient outcomes. Since the reference LDL measurement method, ultracentrifugation is tedious, costly and not suitable for resource-limited settings, the Friedewald formula is one of the most commonly used methods despite its inherent limitations. To estimate LDL-C, the Friedewald equation applies a fixed factor of 5. In contrast, the novel method based on TG and non HDL-C levels uses an adjustable TG: VLDL-C ratio factor. Previous studies attempted to determine LDL using optimal fixed factors [29-31]. For instance, DeLong DM et al., increased the fixed factor from 5 to 6, Hata Y and Nakajima K proposed a higher fixed factor of 5, and Puavilai W et al., proposed a lower fixed factor of 4 [29-31].

In the present study, in group-1 (TG level <100 mg/dL), LDL-C obtained by Martin's formula showed a better correlation with D-LDL than Friedwald's calculated LDL. But at all TG levels, ranging from 101-150 mg/dL, 151-200 mg/dL and 201-400 mg/dL, the Friedewald equation had a slightly better correlation with direct LDL-C in the Indian population. The Friedewald equation was linked to direct LDL-C at all TG levels in the Indian population except for TG levels <100 mg/dL as reported by Krishnaveni P and Gowda VM [32]. However, in the present study, Martin-Hopkins had the least mean difference at TG > 400 mg/dL and suggests Martin's formula may prevent undertreatment due to the underestimation of LDL-C using Friedewald's formula which is consistent with studies done by Kang M et al., and Lee J et al., [33,34]. As per Sirivelu B et al., and Mehta R et al., reports, ROC analysis reconfirmed these findings, i.e., Martin-Hopkins showed better diagnostic performance than

Friedewald's formula [10,35]. The present study results confirmed those Kang M et al., and Lee J et al., [33,34]. When comparing Martin's formula with Friedewald's formula, Farheen F et al., stated that Martin's formula is more accurate, can replace routine Friedewald's formula, and significantly improves LDL-C estimation [12]. The tendency of Friedewald's formula to underestimate LDL-C was also established in the present study.

Limitation(s)

The study on LDL-C estimation has several limitations, including potential bias due to using a calculator without adjusting the factor for the Indian population and the study's subjects not fully representing the general population's baseline characteristics. Furthermore, the need for more data on participants' clinical characteristics or outcomes limits the ability to correlate lipid profiles with overall health or cardiovascular risk. This highlights the need for more comprehensive studies that include diverse population groups and integrate clinical outcomes to ensure the reliability and relevance of LDL-C estimation formulas.

CONCLUSION(S)

In the present study, the Martin-Hopkins formula showed better diagnostic performance among the two equations than Friedewald's formula. The mean difference was lower for Martin's formula than for Friedewald's formula at all TG levels except at levels <100 mg/dL. Considering the mean difference, Martin's formula provides better LDL-C values than Friedewald's formula in the present study's demographic population. Future research should assess the effectiveness of the Martin-Hopkins formula in estimating LDL cholesterol across different populations using comparative studies, direct measurements and new estimation formulas. Updating clinical guidelines, educating healthcare providers, and prioritising patient-centred outcomes are crucial for improving cardiovascular risk assessment accuracy and personalised patient management.

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REFERENCES

[1] Mozaffarian D, Benjamin EJ, Go AS, Arnett DK, Blaha MJ, Cushman M, et al. Heart disease and stroke statistics-2016 update: A report from the American Heart Association. Circulation. 2016;133(4):e38-60.
[2] Stone NJ, Robinson JG, Lichtenstein AH. 2013 ACC/AHA Guideline on the treatment of blood cholesterol to reduce atherosclerotic cardiovascular risk in adults: A Report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines. J Am Coll Cardiol. 2014;63(25):3024-25.
[3] Rosenson RS, Brewer HB, Ansell BJ, Barter P, Chapman MJ, Heinecke JW, et al. Dysfunctional HDL and atherosclerotic cardiovascular disease. Nature Reviews Cardiology. 2016;13(1):48-60.
[4] Friedewald WT, Levy RI, Fredrickson DS. Estimating the concentration of low-density lipoprotein cholesterol in plasma without the preparative ultracentrifuge. Clin Chem. 1972;18(6):499-502.
[5] Dong J, Guo H, Yang R, Li H, Wang S, Zhang J, et al. Serum LDL-and HDL-cholesterol determined by ultracentrifugation and HPLC. JLR. 2011;52(2):383-88.
[6] Martin SS, Giugliano RP, Murphy SA, Wasserman SM, Stein EA, Ceška R, et al. Comparison of low-density lipoprotein cholesterol assessment by Martin/Hopkins Estimation, Friedewald Estimation, and preparative ultracentrifugation: insights from the Fourier trial. JAMA Cardiol. 2018;3(8):749-53.
[7] Miller WG, Myers GL, Sakurabayashi I, Bachmann LM, Caudill SP, Dziekonski A, et al. Seven direct methods for measuring HDL and LDL cholesterol compared with ultracentrifugation reference measurement procedures. Clin Chem. 2010;56(6):977-86.
[8] Martin SS, Blaha MJ, Elshazly MB, Toth PP, Kwiterovich PO, Blumenthal RS, et al. Comparison of a novel method vs. the Friedewald equation for estimating low-density lipoprotein cholesterol levels from the standard lipid profile. JAMA. 2013;310(19):2061-68.
[9] Anandaraja S, Narang R, Godeswar R, Laksmy R, Talwar KK. Low-density lipoprotein cholesterol estimation by a new formula in the Indian population. Int J Cardiol. 2005;102(1):117-20.
[10] Sirivelu B, Namilakonda M, Soma K, Thallapaneni S, Annavarapu DL, Kumar VS, et al. Assessing the validity of nine different formulae for LDL-C estimation in a tertiary care centre, Hyderabad, India. J Clin Diagn Res. 2022;16(1):BC06-BC11.
[11] Johnson R, McNutt P, MacMahon S, Robson R. Use of the Friedewald's formula to estimate LDL-cholesterol in patients with chronic renal failure on dialysis. Clin Chem. 1997;43(11):2183-84.
[12] Farheen F, Ambiger S, Jaalam K, Javali S. Martin's formula as the most suitable method for estimating low-density lipoprotein cholesterol in Indian population. Journal of Laboratory Physicians. 2023;15(04):545-51.
[13] Sampson M, Ling C, Sun Q, Harb R, Ashmaig M, Warnick R, et al. A New equation for calculation of low-density lipoprotein cholesterol in patients with normolipidemia and hypertriglyceridemia. JAMA Cardiol. 2020;5(5):540-48.
[14] Karkhaneh A, Bagherieh M, Sadeghi S, Kheirollahi A. Evaluation of eight formulas for LDL-C estimation in Iranian subjects with different metabolic health statuses. Lipids in Health and Disease. 2019;18(1):231.
[15] Joshi SR, Anjana RM, Deepa M, Pradeepa R, Bhansali A, Dhandania VK, et al. Prevalence of dyslipidemia in urban and rural India: the ICMR-INDIAB study. PLoS One. 2014;9(5):e96808.
[16] Sun T, Chen M, Shen H, Yin P, Fan L, Chen X, et al. Predictive value of LDL/HDL ratio in coronary atherosclerotic heart disease. BMC Cardiovasc Disord. 2022;22(1):273.
[17] Gupta S, Verma M, Singh K. Does LDL-C estimation using Anandaraja's formula better agree with direct LDL-C estimation than Friedewald's? Indian J Clin Biochem. 2012;27(02):127-33.
[18] Agrawal M, Spencer HJ, Faas FH. The method of LDL cholesterol measurement influences the classification of LDL cholesterol treatment goals: A clinical research study. J Investig Med. 2010;58(08):945-49.
[19] Nakanishi N, Matsuo Y, Yoneda H, Nakamura K, Suzuki K, Tatara K. Validity of the conventional indirect methods including Friedewald Method for determining serum low-density lipoprotein cholesterol level: Comparison with the direct homogeneous enzymatic analysis. Journal of Occupational Health. 2000;42(3):130-37.
[20] Vujovic A, Kotur-Stevuljevic J, Spasic S, Bujisic N, Martinovic J, Vujovic M, et al. Evaluation of different formulas for LDL-C calculation. Lipids Health Dis. 2010;9:27.
[21] Lindsey CC, Graham MR, Johnston TP, Kiroff CG, Freshley A. A clinical comparison of calculated versus direct measurement of low-density lipoprotein cholesterol level. Pharmacotherapy. 2004;24(2):167-72.
[22] Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults. Executive summary of the third report of the National Cholesterol Education Program (NCEP) expert panel on the detection, evaluation, and treatment of high blood cholesterol in adults (Adult Treatment Panel III). JAMA. 2001;285(19):2486-97.
[23] National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III). Third Report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III) final report. Circulation [Internet]. 2002;106(25):3143-421.
[24] Grundy SM, Cleeman JI, Merz CN, Brewer HB Jr, Clark LT, Hunninghake DB, et al. Implications of recent clinical trials for the National Cholesterol Education Program Adult Treatment Panel III guidelines. Circulation. 2004;110(2):227-39.
[25] Genest J, McPherson R, Frohlich J, Anderson T, Campbell N, Carpentier A, et al. Canadian Cardiovascular Society/Canadian guidelines for diagnosing and treating dyslipidemia and preventing cardiovascular disease in adults- 2009 recommendations. Can J Cardiol. 2009;25(10):567-79.
[26] Perk J, Backer DG, Gohlke H, Graham I, Reiner Z, Verschuren M, et al. European Guidelines on Cardiovascular Disease Prevention in Clinical Practice (version 2012). The Fifth Joint Task Force of the European Society of Cardiology and Other Societies on Cardiovascular Disease Prevention in Clinical Practice (constituted by representatives of nine societies and by invited experts). Eur Heart J. 2012;33(13):1635-701.
[27] Smith SC Jr, Benjamin EJ, Bonow RO, Braun LT, Creager MA, Franklin BA, et al. AHA/ACCF secondary prevention and risk reduction therapy for patients with coronary and other atherosclerotic vascular disease: 2011 update: An American Heart Association and American College of Cardiology Foundation guideline. Circulation. 2011;124(22):2458-73.
[28] Expert Panel on Detection E. Executive summary of the third report of the National Cholesterol Education Program (NCEP) expert panel on the detection, evaluation, and treatment of high blood cholesterol in adults (adult treatment panel III). JAMA. 2001;285(19):2486-97.
[29] DeLong DM, DeLong ER, Wood PD, Lippel K, Rifkind BM. A comparison of methods for estimating plasma low- and very low-density lipoprotein cholesterol. The lipid research clinics prevalence study. JAMA. 1986;256(17):2372-77. PMID: 3464768.
[30] Puavilai W, Laorugpongse D, Deerochanawong C, Muthapongthavorn N, Srilert P. The accuracy in using modified Friedewald equation to calculate LDL from non-fast triglyceride: A pilot study. J Med Assoc Thai. 2009;92(2):182-87.
[31] Hata Y, Nakajima K. Application of Friedewald's LDL-cholesterol estimation formula to serum lipids in the Japanese population. Japanese circulation journal. 1986;50(12):1191-200.
[32] Krishnaveni P, Gowda VM. Assessing the validity of Friedewald's formula and Anandraja's formula for serum LDL-cholesterol calculation. J Clin Diag Res. 2015;9(12):BC01-04.
[33] Kang M, Kim J, Lee SY, Kim K, Yoon J, Ki H. Martin's equation as the most suitable method for estimating low-density lipoprotein cholesterol levels in Korean adults. Korean J Fam Med. 2017;38(5):263-69.

[34] Lee J, Jang S, Son H. Validation of the Martin method for estimating low-density lipoprotein cholesterol levels in Korean adults: Findings from the Korea National Health and Nutrition Examination Survey, 2009-2011. PLOS one. 2016;11(1):e0148147.

[35] Mehta R, Reyes-Rodríguez E, Bello-Chavolla YO, Guerrero-Díaz AC, Vargas-Vázquez A, Cruz-Bautista I, et al. LDL-C performance was calculated using Martin's formula and compared to the Friedewald equation in familial combined hyperlipidemia. Atherosclerosis. 2018;277:204-10.

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- iThenticate Software: May 24, 2024 (11%)

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- For any images presented appropriate consent has been obtained from the subjects. NA

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