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*Original Article*

# A multidimensional approach to assess cardiovascular disease risk combining biochemical, hematological, lipid ratios, atherosclerotic cardiovascular disease, and WHO/ISH 10-year risk estimators: a cross-sectional study

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**Abstract:** Cardiovascular disease (CVD), a leading global health concern and the primary cause of death worldwide, is often associated with atherosclerosis and cardiac events. Objective: This study evaluated biochemical and hematological parameters as predictors of CVD risk using atherosclerotic CVD and World Health Organization/International Society of Hypertension risk scores (WHO/ISH). Methodology: 102 volunteer participants representing 69 males (67.6%) and 33 females (32.4%) from the Health and Administrative Staff of University Hospital-General Sir John Kotelawala Defence University, Sri Lanka, were selected. Patient demographics, biometrics, clinical parameters, and behavioral risk factors were collected. Laboratory parameters: complete blood count, lipid profile, aspartate and alanine aminotransferases were performed. The 10-year CVD risk was estimated using the "ASCVD-Risk Estimator-Plus" and WHO/ISH risk score charts; South Asia and Southeast Asia. The data were analyzed using IBM-SPSS-version26. Results: The study population showed strong to moderate-strong correlations within the hematological, biochemical, and risk estimators, but not between these parameters. Platelets-to-Lymphocytes Ratio (PLR) had a strong, significant positive correlation with the Neutrophils-to-Lymphocyte Ratio (NLR) and a moderately strong, significant positive correlation with Platelets and NLR ($r=0.446$; $P<0.001$), indicating the inflammatory response, atherosclerosis with increased CVD risk. In the regression analysis, the HDL-LDL ratio was found to independently predict the TC-HDL ratio and successfully combined the WHO/ISH risk estimators with the collected biochemical, hematological, clinical, biometric, and behavioral risk factors by introducing highly predictive, well-fit equations. The ROC analysis indicated that once the HDL-LDL-ratio reduces below 0.39, ASCVD-10-year-risk (ASCVD_10) and ASCVD-lifetime-risk (ASCVD_LT) increase to 4.95 and 37.5, respectively. Conclusion: A strong positive correlation was found between NLR, PLR, and CVD risk, with a moderate correlation between PLT and NLR. ASCVD_10 showed a reliable link with ASCVD_OP and ASCVD_LT, marking the first comparison of all three risk types. Predictive equations were developed by integrating WHO/ISH scores and other parameters. The TC-HDL ratio significantly correlated with the HDL-LDL ratio, enabling a cut-off value to predict ASCVD_10 risk and associated hematological and biochemical markers.

**Keywords:** Cardiovascular diseases, 10-year CVD risk, ASCVD risk estimator, WHO/ISH risk score, NLR, PLR, lipid profile, HDL-LDL ratio

## Introduction

Cardiovascular disease (CVD) remains the leading cause of mortality worldwide, accounting for approximately one-third of all global deaths with 20.5 million deaths reported in 2021 alone [1-4]. "Cardiovascular Diseases" refers to any disease affecting the heart or blood vessels. It is frequently linked to fatty deposits in the arterial walls (atherosclerosis) and an elevated risk of thrombosis [3]. These cardiovascular diseases include myocardial infarction (MI) or ischemic heart disease, stroke, and congestive heart failure. Risk factors for these cardio-

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vascular diseases can be categorized mainly into two groups: controllable factors and uncontrollable factors. Unhealthy diet, physical inactivity, obesity, dyslipidemia, hypertension, diabetes mellitus, smoking, and alcohol consumption can be considered as leading controllable causes. Advanced age, gender, and family history are the major uncontrollable risk factors [3, 5]. Depending on the severity and the progression of the disease, CVDs can be acute or chronic. Acute cardiovascular diseases are characterized by sudden onset and rapid progression. These include myocardial infarction, strokes, and acute heart failure. In such cases, immediate medical intervention is critical to prevent severe complications or death. Chronic cardiovascular diseases, on the other hand, develop slowly over time and include conditions such as hypertension, chronic heart failure, and atherosclerosis. These diseases often persist for months or years, requiring long-term management [5].

Diagnosis for acute CVD events relies on rapid clinical assessments, including electrocardiograms (ECG), cardiac biomarkers (e.g., troponins), and imaging techniques such as echocardiography or angiography. Diagnosis of chronic CVDs is more comprehensive and may involve regular monitoring of blood pressure and cholesterol levels, echocardiography, and stress tests to assess heart function [6]. Several laboratory investigations are requested to diagnose, predict, or determine the prognosis of CVDs. These include complete blood count (CBC) as a routine hematological test, cardiac enzymes, urea/creatinine ratio, serum electrolytes, thyroid function test, liver profile, and brain natriuretic peptides (BNP) [7]. In some cases, D-dimer is also used as a cardiac biomarker for the diagnosis of thrombosis, ischemic heart disease, and cardiovascular mortality [8]. Aspartate Aminotransferase (AST) was the initially used cardiac marker; however, due to its non-specificity for cardiac tissue, it was replaced by other cardiac biomarkers [9].

Lipid profile testing is essential for assessing cardiovascular disease (CVD) risk, as dyslipidemia significantly contributes to atherosclerosis [10, 11]. This test panel includes serum total cholesterol (TC), Low density Lipoprotein cholesterol (LDL), very low-density lipoprotein cholesterol (VLDL), high-density lipoprotein cholesterol (HDL), and triglyceride (TG) [12, 13]. Lipid ratios, such as the total cholesterol to high-density lipoprotein cholesterol (TC-HDL) ratio, high-density lipoprotein cholesterol to low-density lipoprotein cholesterol (HDL-LDL) ratio, and triglycerides to high-density lipoprotein cholesterol (TG-HDL) ratio, have been proposed as superior predictors of cardiovascular events [13-16]. In addition to the lipid parameters, hematological markers, particularly inflammatory indices such as the neutrophil-to-lymphocyte ratio (NLR), platelet-to-lymphocyte ratio (PLR), and monocyte-to-lymphocyte ratio (MLR), have shown strong associations with cardiovascular disease risk [18-23]. These ratios serve as accessible indicators of systemic inflammation, which plays a key role in the development and progression of CVD [17-25].

For accurate risk assessment, risk prediction tools such as the Atherosclerotic Cardiovascular Disease (ASCVD) risk estimator by the American Heart Association and American College of Cardiology and the WHO/ISH 10-Year Risk Charts by the World Health Organization (WHO) in association with the International Society of Hypertension (ISH) have been developed [26-28]. This risk assessment tool has been developed based on the "2013 ACC/AHA Guideline on the assessment of cardiovascular risk" [29, 30]. Age, sex, race, total cholesterol, High-Density Lipoproteins cholesterol (HDL), Low-Density Lipoprotein cholesterol (LDL), systolic blood pressure, diastolic blood pressure, history of diabetes, smoking, treatment for hypertension, and on statins or aspirin therapy are used as parameters for the calculation. This gives three types of atherosclerotic cardiovascular risk for an individual: 10-year ASCVD risk (ASCVD_10), Lifetime ASCVD risk (ASCVD_LT), and Optimal ASCVD risk (ASCVD_OP) [28].

Similarly, WHO/ISH Cardiovascular Risk Estimator charts have been developed for different regions in the world. In this study, mainly Southeast Asia (Sri Lanka, Indonesia, Cambodia, Lao PDR, Maldives, Myanmar, Malaysia, Philippines, Thailand, Timor-Leste, Vietnam, Mauritius, Seychelles) and South Asia (Bangladesh, Bhutan, India, Nepal, Pakistan) risk charts have been used to evaluate the CVD risk. The risk is expressed as percentages in laboratory-based and non-laboratory-based charts. Laboratory-based risk charts can be used when laboratory testing is readily available, and non-laboratory-based risk charts can be used where laborato-

ry testing is limited. This clinical tool assesses cardiovascular events, including myocardial infarction, stroke, or cardiovascular death, in adults aged 40-75 years. This estimator computes the individuals' risk for the development of CVD based on age, gender, blood pressure (mmHg), cholesterol levels (mmol/l), smoking status, and diabetes. The WHO/ISH Cardiovascular Risk Estimator tool is based on data from multiple populations, including East Asia, Central Asia, South Asia, etc., and considers the specific risk factors and the cut-off values for each region. As this tool provides an estimation of the 10-year risk of developing cardiovascular diseases, it assists healthcare providers in identifying individuals at high risk. Those individuals can benefit from preventive interventions, such as lifestyle modifications and pharmacological treatments. The tool offers a personalized approach to risk stratification, based on different global contexts. This estimator is useful in decision-making about treatment and management in primary care settings [31-34].

However, while these traditional variables provide a general risk assessment, they may not capture the full complexity of cardiovascular disease in certain populations or individuals. The use of additional variables in risk assessment improves predictive accuracy [35, 36]. Therefore, incorporating specific biochemical and hematological parameters into cardiovascular risk prediction models would allow for a more comprehensive assessment by accounting for factors such as inflammation and lipid-related risks, which current models do not fully capture. The development of an accessible and cost-effective predictive marker represents a pivotal advancement in reducing preventable CVD fatalities. Existing research has examined the role of clinical, laboratory and behavioral risk factors in cardiac risk assessment. Nevertheless, there is a notable absence of studies focusing on combining these previously mentioned parameters and existing risk estimators to determine the CVD risk prediction accurately.

This study was motivated by the critical need to enhance CVD risk prediction by integrating biochemical and hematological parameters with established risk estimators tailored to South Asian and Southeast Asian populations. By combining traditional risk factors with routinely tested laboratory markers, this study aims to develop a more comprehensive, cost-effective and clinically applicable predictive model. This focuses on the primary prevention of CVD and has the potential to identify individuals at hidden risk who appear healthy but suffer from subclinical disease. This will be useful to initiate preventive measures that potentially reduce the development of CVD, promote cardiovascular health awareness, and be cost-effective in the long term by reducing the burden of acute cardiovascular events. Further, these findings are expected to contribute to better CVD health awareness and management, particularly in resource limited settings.

## Materials and methods

This study was a prospective cross-sectional study. Ethical clearance was granted by the Ethical Review Committee of General Sir John Kotelawala Defense University (ERC No: RP/S/2022/11). The sample size was calculated using Cochran's formula, considering 1.96 as the standard normal deviation for a 95% confidence level, 6.9% (0.069) as the expected proportion in the population based on Jayawardene et al. 2017, and 0.05 as the precision. Substituting these values into the formula: 102 volunteer participants of the health and administrative staff of University Hospital-KDU (UH-KDU) between 40 to 70 years were selected for the study, and all the participants were Sri Lankans (South Asians). The selection was based on convenient sampling, facilitating the timely collection and analysis of the samples and ensuring efficient data gathering. Exclusion criteria included a history of CVDs (Myocardial infarction, coronary artery bypass graft surgery, percutaneous coronary intervention) to avoid cofounding effects of existing CVD on study outcomes. Participants currently on cholesterol-lowering drugs (statins) and antiplatelet drugs (aspirin, clopidogrel) were excluded because these medications directly affect lipid profiles and blood parameters. Individuals with non-cardiac diseases that affect the CBC, lipid profile and liver profile were excluded to reduce confounding biological variabiity. Pregnant or lactating women were excluded due to physiological changes that alter blood and lipid parameters. This study has selected a CVD risk group focusing on primary prevention. Risk estimators, such as the ASCVD risk calculator and WHO/ISH risk assessment charts, are particularly effective in predicting the 10-year risk of

developing CVD and are best applied to low-risk groups to identify individuals who may benefit from early interventions. The investigators conducted the initial screening of the study participants, and their eligibility to participate in the study according to the inclusion and exclusion criteria was confirmed by a consultant cardiologist at UHKDU. The blood samples will be obtained from the participants only after their eligibility for the study is confirmed and their consent is obtained.

*Laboratory analysis*

Following a 10-12 hour fasting period, the blood samples were collected into Ethylene Diamine Tetra Acetic acid (EDTA) & plain blood collection tubes by a qualified nurse. CBC Lipid profile parameters of the participants were performed manually (Spectrum Diagnostics and Healthcare, Bangalore, India) using a spectrophotometer (Double Beam UV/Vis Spectrophotometer CT-2300). For the TC estimation, the Cholesterol Oxidase Peroxidase 4-Aminoantipyrine method (CHOD-PAP) was followed, while Glycerol Phosphate Oxidase-Peroxidase (GPO-PAP) was carried out for the TG estimation. HDL cholesterol differential precipitation method was performed to obtain the HDL concentrations. LDL level was calculated by the "Friedewald equation". TC-HDL & HDL-LDL ratios were derived by calculations. Internal quality controls and the standards were performed using the quality control and standard reagents purchased with the same reagent kit. The AST & Alanine aminotransferase (ALT) assays were performed by the automated biochemistry analyzer Selectra Pro-XL (Gesan Production, 71 Via Fiera Dell'eremita, Campobello Di Mazara, Sicilia 91021, Italy), while the AST/ALT ratio was obtained through calculations. The Complete Blood Count (CBC) was carried out by automated hematology analyzer Sysmex XP-100 and cross-checked by performing manual differential counts. Parameters such as total WBC count, hemoglobin, hematocrit & PLT count were obtained though the analyser. Two inflammatory indices were derived from the above parameters, NLR; by deviding the absolute neutrophil count by absolute lymphocyte count, & PLR; by deviding absolute platelet count by absolute lymphocyte count. The ASCVD risk was estimated by the "ASCVD Risk Estimator Plus-Mobile and Web App" developed by the American College of Cardiology and the American Heart Association. If the score for 10-year ASCVD risk falls between 0-4.9%, it is considered as 'low risk', 5-7.4% means 'borderline risk', 7.5-20% means 'intermediate risk' and >20% implies 'high risk'. For lifetime ASCVD risk, the cut-off value is 39; high risk ≥39% and low risk <39%. The ASCVD_OP provides the optimal risk levels for lifetime risk. The optimal conditions are defined as follows: untreated total cholesterol (TC) less than 180 mg/dL, untreated blood pressure less than 120/80 mmHg, no history of diabetes, and non-smoking status. The patient's risk is calculated by comparing their current parameters to these optimal conditions. The WHO/ISH CVD risk estimator chart can distinguish an individual into low (<10%), moderate (10-20%), high (20-30%), and very high (>30%) risk groups.

*Statistical analysis*

Results of the biochemical and hematological parameters were statistically analyzed by a computerized database using Microsoft Excel and SPSS version 26 (IBM Corp., 2017, IBM SPSS Statistics for Windows, Armonk, NY). First, the univariate analysis was performed by checking the normality of individual data sets according to the Kolmogorov-Smirnov normality check. P>0.05 was considered as the normal distribution for the data. Since certain categories of data were not in the normal distribution, the analysis was confined to both parametric and non-parametric. The mean, median and standard deviation (SD) were calculated for each laboratory investigation using SPSS. In the multivariate analysis, the Correlation bivariate analysis was first performed to establish correlations between the blood parameters. Pearson's Bivariate analysis was used when the two sets of data were in the normal distribution. In comparison, the Spearman Bivariate analysis was used when two sets of data or at least one set of data, did not follow the normal distribution. As the study parameters clearly consist of three types: hematological parameters (NLR & PLR), biochemical parameters (AST, ALT, AST/ALT ratio, TC, TG, HDL, LDL, TC-HDL ratio & HDL-LDL ratio), and ASCVD risk types (ASCVD-10, ASCVD-LT & ASCVD-OP). Linear Regression analysis was performed to predict the HDL/LDL ratio and other Cardiovascular Risk estimators (as dependent parameters) with other hematological and biochemical investigations (as independent parameters) within the

above-mentioned groups. Finally, the receiver-operating characteristic (ROC) curve analysis was used to determine the optimum cut-off level of the hematological and biochemical parameters in association with the HDL-LDL ratio. The Kruskal-Wallis & Mann-Whitney U tests analysis was performed to determine the significance of the mean values of HDL in different groups of LDL from normal to high range. Statistical significance was defined as $P<0.05$.

## Results

The study category consisted of 102 participants, representing 69 males (67.6%) and 33 females (32.4%) between 40 and 70 years old, with an age mean of 49.22±9.02. The mean SD values of the biochemical test results, AST, ALT, AST/ALT ratio, TC, TG, HDL, LDL, TC-HDL ratio, HDL-LDL ratio, some of the hematological parameter results, NLR, PLR, and the calculated results of the risk estimators were tabulated (**Table 1**). By observing the means (**Table 1**), most values were in the normal range. However, the LDL value is near optimal (100-129 mg/dl), and the mean systolic and diastolic blood pressures are slightly higher than the reference ranges. The mean ASCVD-10 risk estimator for the selected population falls within the borderline risk category, whereas the ASCVD-LT mean suggests a low-risk [26-28]. In accordance with the whole category, the male category (n=69) also showed similar deviations, but it is comparatively high. When considering the mean values of the same parameters in the female category (n=33), TC was slightly elevated, LDL levels were borderline high and the ASCVD_10 is low, while the ASCVD_LT is high, though it falls just above the cut-off for this category.

*Normalization of the data*

In the first step, the data of the whole male and female categories were tested for normality. In the whole category, out of all the parameters, WBC, HDL, LDL, TC-HDL ratio, and HDL-LDL ratio showed a normal distribution ($P>0.05$). The other parameters, NLR, PLR, AST, ALT, AST-ALT ratio, TC, TG, ASCVD_10, ASCVD_LT, ASCVD_OP and WHO/ISH 10-year risk score did not follow a normal distribution ($P<0.05$). When considering the normality results of the male and female categories, only the TC, HDL and LDL could follow normality in both groups.

To find out whether there was any significant difference between males and females in said parameters, the parametric (Independent T-test) for TC, HDL, LDL and the non-parametric (Mann-Whitney U) test were carried out. None of the tests were able to provide a significant ($P<0.05$) result. Therefore, the rest of the analysis was confined to the whole group, not to males and females separately.

*Correlation bivariate analysis*

In the whole group (n=102), the Pearson/Spearman bivariate analysis of the data was carried out depending on the normality. Strong to moderate-strong correlations were observed only among the same type of parameters, i.e., either among hematological, biochemical or risk estimators. The PLR had a strong, significant positive correlation with the NLR ($r=0.700$; $P<0.001$), while the PLT had a moderately strong significant positive correlation with the NLR ($r=0.446$; $P<0.001$). HDL-LDL ratio provided several strong to moderately strong negative and positive correlations with their adjacent parameters of TC-HDL ratio ($r=-0.979$; $P<0.001$), TC ($r=-0.589$; $P<0.001$), LDL ($r=-0.825$; $P<0.001$), and HDL ($r=0.529$; $P<0.001$). In Risk estimators, the ASCVD_10 provided significant strong positive to moderately strong positive correlations with the ASCVD_OP ($r=0.701$; $P<0.001$) and ASCVD_LT ($r=0.647$; $P<0.001$). The WHO 10 years Risk estimator too provided significant strong positive to moderately strong positive correlations with the ASCVD_10 ($r=0.829$; $P<0.001$) ASCVD_OP ($r=0.647$; $P<0.001$) and ASCVD_LT ($r=0.522$; $P<0.001$). No reliable correlations were observed between the hematological, biochemical and parameters or risk estimators.

*Receiver Operating Characteristic (ROC) curve analysis*

Finally, the Receiver Operating Characteristic (ROC) curve analysis was performed to find a cut-off value for the HDL-LDL ratio of 0.39. The two groups were made as; Group 1: HDL-LDL ratio ≤0.39, Group 2: HDL-LDL ratio >0.39. In the ROC analysis, a few hematological and biochemical parameters were able to provide cut-off values to predict HDL-LDL ratio for 0.39 cut-off level with significant moderate to high sensitivity, specificity, and Area Under Curves (AUC ≥0.600) (**Figure 1** and **Table 2**).

**Table 1.** Mean, median and standard deviation (SD) values of the laboratory parameters of whole, male, and female categories used in the study

| Category | All (n=102) | | | Male (n=69) | | | Female (n=33) | | |
|---|---|---|---|---|---|---|---|---|---|
| Descriptive statistics | Mean | Median | SD | Mean | Median | SD | Mean | Median | SD |
| AST (IU/L) | 30.5 | 27.8 | 13.2 | 32.9 | 29.1 | 14.2 | 25.4 | 22.1 | 8.9 |
| ALT (IU/L) | 44.4 | 31.3 | 27.9 | 46.6 | 39.4 | 30.4 | 27.3 | 23.0 | 15.4 |
| TC (mg/dl) | 197.5 | 191.1 | 37.2 | 193.2 | 189.8 | 32.8 | 205.0 | 202.0 | 44.5 |
| TG (mg/dl) | 105.6 | 94.7 | 45.6 | 110.3 | 99.0 | 49.0 | 95.9 | 83.5 | 36.4 |
| HDL (mg/dl) | 46.6 | 46.8 | 10.1 | 46.4 | 45.4 | 11.6 | 47.1 | 48.3 | 6.1 |
| LDL (mg/dl) | 128.8 | 126.2 | 35.2 | 124.7 | 122.3 | 31.6 | 137.5 | 128.0 | 40.6 |
| TC-HDL ratio | 4.39 | 4.15 | 1.12 | 4.39 | 4.02 | 1.24 | 4.36 | 4.20 | 0.81 |
| HDL-LDL ratio | 0.39 | 0.37 | 0.15 | 0.40 | 0.36 | 0.17 | 0.37 | 0.34 | 0.11 |
| WBC ($\times 10^9$/L) | 7.07 | 6.75 | 2.08 | 7.09 | 6.80 | 2.04 | 7.00 | 6.60 | 2.18 |
| NLR | 1.40 | 1.15 | 0.72 | 1.35 | 1.09 | 0.70 | 1.51 | 1.47 | 0.74 |
| PLR | 106.4 | 102.5 | 35.3 | 102.0 | 94.9 | 37.0 | 115.6 | 115.1 | 29.7 |
| SYS/DIA BP (mmHg) | 128.7/84.4 | 129.0/85 | 13.9/10.7 | 128.2/84.4 | 129.0/85.0 | 12.7/10.1 | 129.7/84.2 | 128/84 | 16.2/11.9 |
| ASCVD_10 (%) | 5.94 | 3.55 | 5.83 | 6.61 | 4.10 | 6.46 | 4.52 | 2.70 | 3.92 |
| ASCVD_LT (%) | 46.84 | 50.00 | 10.99 | 49.89 | 50.00 | 10.09 | 40.50 | 39.00 | 10.19 |
| ASCVD_OP (%) | 2.16 | 1.05 | 2.70 | 2.36 | 1.00 | 3.07 | 1.73 | 1.10 | 1.64 |

Abbreviations mentioned in the table are as follow: AST, Aspartate aminotransferase; ALT, Alanine aminotransferase; TC, Total Cholesterol; TG, Triglyceride; HDL, High density Lipoproteins; LDL, Low-density lipoproteins; TC-HDL, Total cholesterol: HDL ratio; NLR, Neutrophils to Lymphocytes ratio; PLR, Platelets to Lymphocyte ratio; SY_BP, Systolic blood pressure; DIA_BP, Diastolic blood pressure; ASCVD_10, Atherosclerotic Cardiovascular Diseases-10 year risk; ASCVD_LT, Atherosclerotic Cardiovascular Diseases-Life time risk; ASCVD_OP, Optimal Atherosclerotic Cardiovascular Diseases risk.

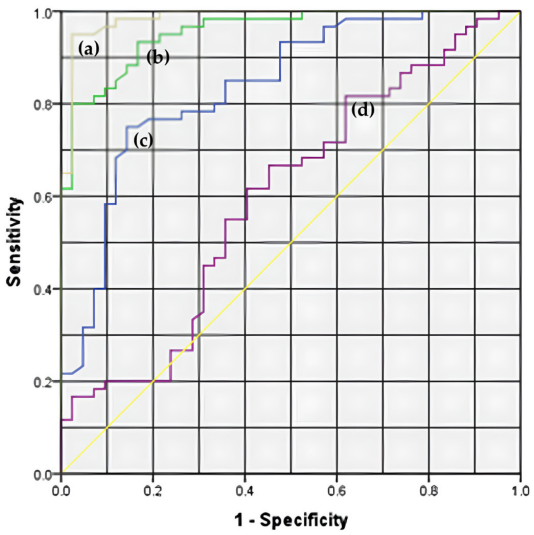


**Figure 1.** Receiver Operating Characteristic (ROC) curve analysis of a few hematological and biochemical parameters to predict HDL: LDL ratio (≤0.39); (a) TC: HDL ratio, (b) LDL, (c) TC, (d) PLR.

In addition, the same cut-off value of HDL-LDL ratio (0.39) was used to predict the risk estimator for ASCVD_10 and ASCVD_LT with high sensitivity. The ASCVD_OP did not provide reliable results. The details are enclosed in **Figure 2** and **Table 2**.

The data reveal that the cut-off values (**Table 2**) of a few hematological and biochemical parameters have been derived with high sensitivity and/or high specificity, with an AUC >0.600.

The results of **Table 2** indicate that once the HDL-LDL ratio reduces below 0.39, the cut-off values of the parameters will increase above the stated levels. The ROC curve analysis was further extended based on the ASCVD_10 risk group categories: Group 1: ASCVD_10 ≤5.0%, Group 2: ASCVD_10 >5.0%. The ROC curve analysis results showed that the cut-off value for HDL-LDL is 0.40 with a sensitivity of 55.0%, specificity of 81.0% and Area Under Curve (AUC) of 0.709 (**Figure 3**).

*Non-parametric analysis by different LDL groups*

Since both LDL and HDL were in the upper normal and lower normal ranges, respectively, the behavior of the HDL was tested using the same data in different groups of LDLs, from normal to the high range. The 4 groups were designed according to the classifications given in literature [37]; Group 1: LDL ≤100 mg/dl (optimal), Group 2: 100 mg/dl < LDL ≤128 mg/dl (near-optimal), Group 3: 128 mg/dl < LDL ≤159 mg/dl (borderline high), Group 4: LDL >159 mg/dl (High). In both Kruskal-Wallis & Mann-Whitney U tests analysis showed no significant difference in HDL or TG between any of the aforesaid LDL groups among all. The other parameters: TC, HDL-LDL ratio and TC-HDL ratio showed significant mean differences (P<0.01) between the two groups. The mean values of the lipid parameters used in the Kruskal-Wallis & Mann-Whitney U test analysis are shown in **Table 3**.

According to the Mann-Whitney U tests analysis of the parameters in **Table 3**, the mean HDL values slightly decrease while the LDL levels increase but do not significantly (P>0.05) vary among groups. Both TC and TC-HDL values increase with rising LDL levels. However, the TG values do not change significantly (P>0.05) across the four groups.

Cardiovascular risk was estimated using the World Health Organization (WHO) cardiovascular risk charts developed for the South Asian population (Including India, Pakistan, Bangladesh, and Nepal). Although Sri Lanka is not included in this chart or does not have a specific CVD chart developed for its population, this chart can still be effectively used to assess CVD risk as it represents the majority of South Asian countries. These charts are developed to estimate the 10-year risk of CVD for individuals in different regions, considering the specific risk factors and the cut-off values for each region. Charts are available in two formats: laboratory-based risk chart, which can be used when laboratory testing is readily available, and non-laboratory-based risk chart can be used where laboratory testing is limited. In this study, laboratory-based risk estimation was considered for the CVD risk estimation. Laboratory-based risk charts are further stratified based on the diabetic status, age, gender, systolic blood pressure value, smoking status, and total cholesterol level. This study used the laboratory-based risk chart to estimate the 10-year CVD risk. In the risk assessment chart, risk levels are categorized by percentage ranges <5%, 5% to <10%, 10% to <20%, 20% to <30%, and ≥30% [38].

*Regression analysis*

In the regression analysis (n=102), the HDL-LDL ratio was found to predict the TC-HDL ratio independently:

**Table 2.** Results of the ROC curves used to predict HDL-LDL ratio from a few hematological and biochemical parameters and predict risk estimators

| Parameter | The cut-off value for HDL-LDL ratio# of 0.39 | Sensitivity (%) | Specificity (%) | AUC | Sig. (p) |
|---|---|---|---|---|---|
| TC-HDL ratio | 3.96 | 95.0 | 99.7 | 0.986 | 0.000 |
| LDL | 112.2 | 93.3 | 83.3 | 0.956 | 0.000 |
| TC | 191.1 | 75.0 | 85.7 | 0.838 | 0.000 |
| PLR | 96.2 | 66.7 | 54.8 | 0.599 | 0.089 |
| ASCVD_10 | 4.95 | 46.0 | 92.7 | 0.762 | 0.081 |
| ASCVD_LT | 37.5 | 90.0 | 25.0 | 0.611 | 0.064 |

#Cut-off values were obtained based on two HDL-LDL ratio groups; Group1: HDL-LDL ratio ≤0.39; Group2: HDL-LDL ratio >0.39. The values of the parameters were increasing with the decreasing levels of HDL-LDL ratio.

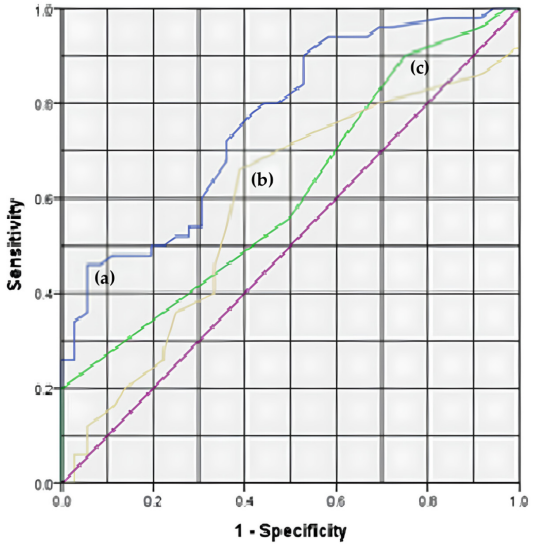


**Figure 2.** Receiver Operating Characteristic (ROC) curve analysis to predict risk estimator for HDL-LDL ratio (≤0.39); (a) ASCVD_10, (b) ASCVD_LT, (c) ASCVD_OP.

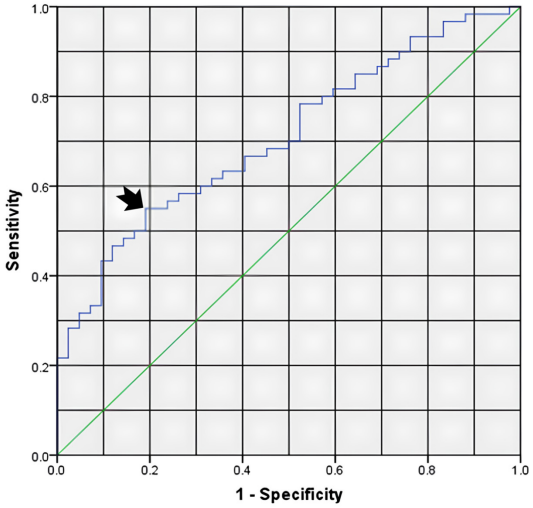


**Figure 3.** Receiver Operating Characteristic (ROC) curve analysis to predict HDL-LDL ratio from the ASCVD_10 (>5.0).

HDL-LDL ratio = 1.2 * (TC-HDL ratio) + 0.921 (1)

$R^2$=0.769; Sig. TC-HDL ratio P=0.07 & Sig. Constant P=0.03; VIF=1.000; Durbin-Watson (DW) statistic test value =1.681.

The regression analysis has been extended to predict the risk estimators from the biochemical and hematological parameters used in the study.

According to the Southeast Asia (SEA) WHO risk estimation chart, the derived equation of the best among all the combinations is as follows:

WHO 10-year risk level (SEA) = 0.310 * (ASCVD_10) + 0.196 * (Age) + 1.552 * (Smoking status) + 2.360 * (Diabetes status) + 0.051 * (Systolic Blood Pressure) -13.786 (2)

$R^2$=0.915; Adjusted R2=0.911; Sig. F Change =0.000.

Sig. and variance inflation factors (VIF) values; ASCVD_10 (P=0.000, VIF=3.142), Age (P=0.000, VIF=2.659), Smoking status (P=0.000, VIF=1.568), Diabetes status (P=0.000, VIF=1.176), Systolic Blood Pressure (P=0.000, VIF=1.093), Constant (P=0.000), DW statistic test value =1.926.

According to the South Asia (SA) WHO risk estimation chart, the derived equation of the best among all the combinations is as follows:

WHO 10-year risk level (SA) = 0.337 * (ASCVD_10) + 0.184 * (Age) + 1.897 * (Smoking status) + 2.470 * (Diabetes status) + 0.043 * (Systolic Blood Pressure) + 0.006 * (TC) + 0.038 * (HDL) -14.807 (3)

**Table 3.** Mean values of the Lipid Profile Parameters in different LDL Groups

| Group | Description | HDL (Mean) | TC (Mean) | TG (Mean) | HDL-LDL ratio (Mean) | TC-HDL ratio (Mean) |
|---|---|---|---|---|---|---|
| 1 | LDL ≤100 mg/dl (optimal) | 47.7 | 157.2 | 108.0 | 0.55 | 3.39 |
| 2 | 100 mg/dl < LDL ≤128 mg/dl (near optimal) | 47.6 | 185.9 | 105.1 | 0.41 | 4.07 |
| 3 | 128 mg/dl < LDL ≤159 mg/dl (borderline high) | 45.2 | 210.5 | 104.7 | 0.31 | 4.86 |
| 4 | LDL >159 mg/dl (High) | 45.6 | 244.3 | 104.6 | 0.26 | 5.48 |

$R^2$=0.928; Adjusted R2=0.923; Sig. F Change =0.000.

Sig. and VIF values; ASCVD_10 (P=0.000, VIF= 3.445), Age (P=0.000, VIF=2.750), Smoking status (P=0.000, VIF=1.612), Diabetes status (P=0.000, VIF=1.195), Systolic Blood Pressure (P=0.000, VIF=1.101), TC (P=0.055, VIF= 1.104), HDL (P=0.002, VIF=1.166), Constant (P=0.000), DW statistic test value =1.881.

Further, an equation was derived without considering ASCVD_10 as follows:

WHO 10-year risk level (SA) = 0.352 * (Age) + (-1.950) * (Gender) + 3.177 * (Smoking status) + 3.983 * (Diabetes status) + 0.068 * (Systolic Blood Pressure) + 0.015 * (TC) -24.236 (4)

$R^2$=0.895; Adjusted R2=0.888; Sig. F Change =0.000.

Sig. and VIF values; Age (P=0.000, VIF=1.130), Gender (P=0.000, VIF=1.403), Smoking status (P=0.000, VIF=1.162), Diabetes status (P= 0.000, VIF=1.245), Systolic Blood Pressure (P= 0.000, VIF=1.052), TC (P=0.000, VIF=1.029), Constant (P=0.000), DW statistic test value =2.044.

None of the ASCVD Risk estimators were able to find reliable regression equations as the predictors for the other parameters.

The B values (regression coefficients) for each parameter were extracted from the risk equations (Equations 2 and 4) and were plotted in a graph (**Figure 4**) to compare their strength as independent values for the said two risk estimators. Parameters were categorized based on their presence in one or both equations (TC and HDL were only present in the South Asian chart). The R Square values and the DW statistic test values were also plotted in the same graph to compare the strength of the equations derived by SPSS.

## Discussion

"Cardiovascular disease" (CVD) is a leading cause of death worldwide, which is often linked to atherosclerosis and thrombosis [4, 39]. Accurate risk assessment is crucial for prevention and to guide the use of statins, aspirin, and other interventions. This study implements a multidimensional approach to cardiovascular risk assessment, integrating hematological and biochemical parameters along with ASCVD and WHO/ISH 10-year risk estimators. By incorporating routinely performed, cost-effective laboratory tests together with biometric, clinical, and behavioral risk factors, this model offers a comprehensive approach for enhancing early detection and prevention of CVD. **Table 1** reveals that the whole category (n=102) has distinct SDs of most of the parameters from their respective means other than the ratios; HDL-LDL and TC-HDL and the male category followed it. The lesser deviations in HDL-LDL and TC-HDL ratios indicate that these lipid levels are relatively stable within the selected sample population. This consistency suggests that for this cohort, there might be uniformity in factors affecting lipid metabolism, such as diet and physical activity [40]. A similar study reported low lipid parameter variability among a rural Indian population where uniform physical activity levels and dietary habits were observed [41]. The mean values obtained for each parameter were consistent with similar studies conducted in Sri Lanka [42, 43]. Mean LDL values of males, females, and the entire population were near optimal (100 mg/dL < LDL ≤128 mg/dL). Research has shown that Asians tend to have higher LDL levels compared to Western populations [44].

At the first step of the statistical analysis, the data of each parameter was tested for normality. Since the majority of the parameters were not in a normal distribution, most of the multivariate analysis had to be carried out as non-

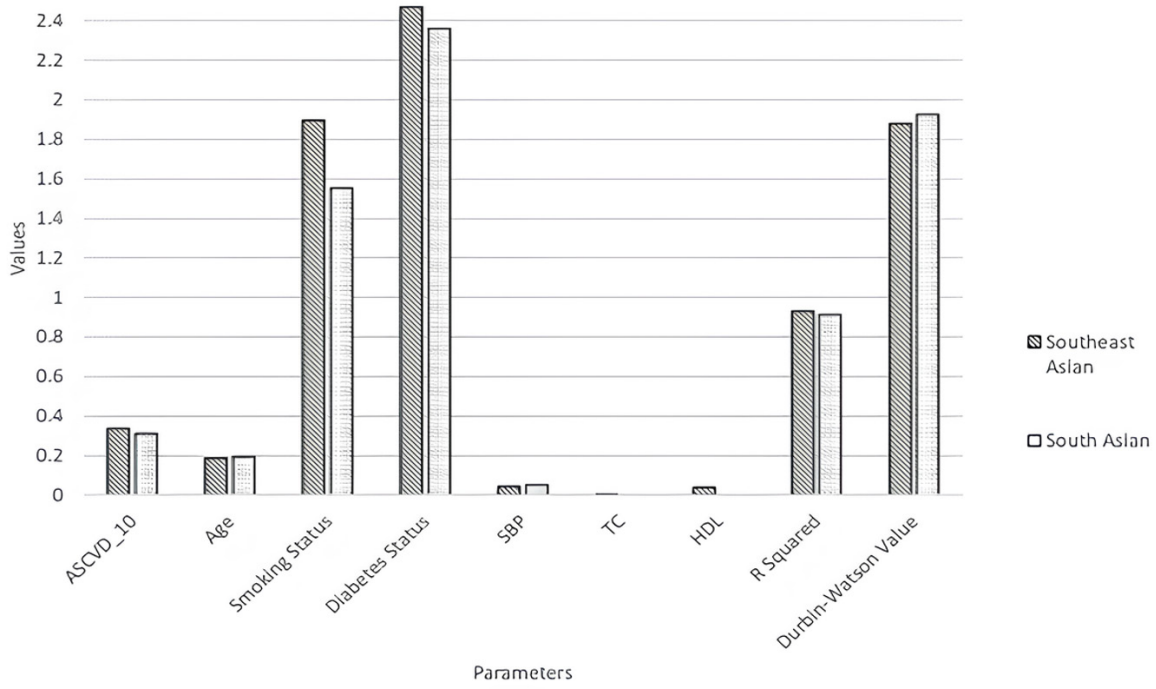


**Figure 4.** Comparison of regression coefficients (B values), $R^2$ values, and DW statistic test values for paramet ers contributing to ASCVD 10-year risk equations in Southeast Asian and South Asian risk charts (SBP = systolic blood pressure; TC = Total cholesterol; HDL = High-density lipoproteins).

parametric. In Spearman's correlation bivariate analysis, strong to moderately strong correlations were observed only among the same type of parameters, i.e., among hematological, biochemical, or risk estimators. Further, results revealed that PLR had a strong, significant positive correlation with the NLR (r=0.700; P< 0.001). In recent studies, researchers have estimated the correlation between NLR and PLR separately with CVD risk, examining their independent effects and the focus has shifted from considering the single parameters to the use of parameter ratios for more comprehensive analysis. A study carried out on the prognostic value of hematological parameters participating in patients with acute myocardial infarction revealed that NLR acts as an independent predictor of in-hospital mortality and has a strong correlation to CVD [45, 46]. Many studies have demonstrated an association between PLR and adverse cardiac events and mortality [47, 48]. Korkmaz et al. (2015) and Larman et al. (2020) revealed that NLR and PLR had elevated values in CVD and CVD outcomes compared to normal. Moreover, a moderately strong significant positive correlation with PLT and NLR (r=0.446; P<0.001) indicates the increased CVD risk. These hematological parameters were commonly studied as inflammatory markers used as prognostic factors in various diseases, including CVDs [26, 29].

In this study, the HDL-LDL ratio provided several strong to moderately strong negative and positive correlations with their adjacent parameters of TC-HDL ratio (r=-0.979; P<0.001), TC (r=-0.589; P<0.001), LDL (r=-0.825; P<0.001), and HDL (r=0.529; P<0.001). Several studies have shown that the TC-HDL ratio is a strong cardiovascular risk marker [17, 49-51]. Various research groups have recently investigated the relationship between HDL-LDL ratio and CVDs. A study conducted by Yuan et al. (2023) for the investigation of the correlation of HDL-LDL ratio with MI, all-cause mortality, hemorrhagic stroke, and ischemic stroke reveals that the ratio of 0.4-0.6 correlates with lower risk, while the ratio <0.4 correlates with higher risk for the selected CVDs [52]. Similarly, this study also successfully obtained the HDL-LDL cutoff value of 0.39 by analysis of ROC curves. Gowtham et al. (2012) imply that the HDL-LDL ratio could be used as a cardiovascular matter in type 2 diabetics instead of the lipoprotein(a), stating that the HDL-LDL ratio is a more practi-

cal and inexpensive choice than Lp(a) estimation [53].

Moreover, the ASCVD_10 has significant strong positive to moderately strong positive correlations with ASCVD_OP (r=0.701; P<0.001) and ASCVD_LT (r=0.647; P<0.001). The correlation between ASCVD_10 and ASCVD_LT was consistent with findings from previous research [12]. To our knowledge, no reported studies have compared all three risk estimators in one study, making these findings valuable to existing research. These correlations highlight the interconnectedness of short-term and long-term cardiovascular risks, underscoring the importance of integrating both assessments in clinical practice to guide comprehensive preventive strategies. However, any significant correlations between the hematological and biochemical parameters or the risk estimators were not observed in this study. In the regression analysis, the HDL-LDL ratio was predicted independently from the TC-HDL ratio as, HDL-LDL ratio = 1.2 * (TC-HDL ratio) + 0.921 with a reliable R2 (0.769). This finding suggests a robust linear relationship between these lipid ratios, highlighting the potential for the TC-HDL ratio to predict the HDL-LDL ratio. The regression equation indicates that for every unit increase in the TC-HDL ratio, the HDL-LDL ratio increases by 1.2 units, with a baseline offset of 0.921 (approximately 1.0). This finding could simplify cardiovascular risk prediction by allowing us to estimate the HDL-LDL ratio directly from the TC-HDL ratio that is being extensively used, thus providing a more streamlined approach to lipid management. These results align with previous studies that have identified the importance of lipid ratios in predicting cardiovascular risk. Similar research emphasized that lipid ratios are superior to individual lipid parameters in assessing risk [51-57]. Calling et al. (2019) revealed that the TC-HDL ratio substantially predicts acute MI in middle-aged women [57]. This indicates that it should be included in risk assessment tools to facilitate early detection. In a middle-aged male population, an elevated serum LDL-HDL ratio was independently linked to an elevated risk of sudden cardiac death, but not the individual lipid parameters [58]. These findings confirm the significance of the TC-HDL ratio in effectively predicting the HDL-LDL ratio; this study supports the continued use of these ratios in clinical practice and risk stratification.

WHO/ISH 10-year CVD risk estimation charts are an effective framework for evaluating the probability of an individual developing a major cardiovascular event within a decade. These charts have been developed for different regions of the world. In this study, mainly Southeast Asia (SEA), including Sri Lanka and South Asia (SA) risk charts, have been used to evaluate the CVD risk. These charts have been prepared considering a comprehensive set of parameters, including age, gender, blood pressure, cholesterol levels, smoking habits, and diabetes status. This is particularly significant in the Asian demographic, where the rising incidence of CVD is exacerbated by rapid urbanization, evolving dietary habits, and genetic predispositions to diabetes and hypertension [3, 59].

This study developed three regression equations (Equations 2-4) for predicting the WHO/ISH 10-year CVD risk level considering the SEA and SA risk charts. The developed equations were statistically significant (P=0.000) and achieved a high coefficient of determinations (Equation 2; $R^2$=0.915, Equation 3; $R^2$= 0.928, Equation 4; $R^2$=0.895) and an adjusted $R^2$ values of (Equation 2; Adjusted R2=0.911, Equation 3; Adjusted R2=0.923, Equation 4; Adjusted R2=0.888) indicating a good fit of the model and high accuracy.

In Equation 2, the Southeast Asian population was considered, incorporating key predictors such as ASCVD_10 risk, age, smoking status, diabetes status, and systolic blood pressure. The model's high $R^2$ (0.915) and adjusted $R^2$ (0.911) indicate strong predictive power, while the statistical significance of all variables (P= 0.000) and the absence of multicollinearity (VIF <5) underscore its reliability. Equally, the presence of a DW statistic test value closer to 2.0 indicates the model is free of autocorrelations. Notably, the substantial weights for diabetes status and smoking status highlight the critical role these factors play in cardiovascular risk.

In the third equation, only the South Asian population was considered. Each predictor variable contributes significantly to the model, as evidenced by their respective *p*-values. Age, smoking status, diabetes status, SBP, and HDL cholesterol are statistically significant (P<0.05), emphasizing their critical roles in CVD risk prediction. Notably, TC approaches significance

(P=0.055), suggesting it may still hold clinical relevance in larger samples or different populations. The variance inflation factors (VIFs) for all variables are below 5, with most were under 2, indicating no multicollinearity issues. The Durbin-Watson statistic of 1.881 suggests that residuals are independent, further supporting the robustness of the model. By further studying the developed model, smoking and diabetes status contributed the most, while other parameters provided minimal contribution, even though they fit well into the model. ASCVD_10 risk score, too, made a substantial contribution to the model.

Several studies have examined how well the WHO/ISH 10-years CVD risk estimation tools perform, especially within Asian populations. A review conducted in 2022 evaluated several CVD risk calculators, including the WHO/ISH risk scores. The findings revealed that tools such as QRISK® demonstrated higher accuracy for certain groups, whereas the WHO/ISH scores were among the least reliable. This review underscored the importance of creating and validating risk prediction models that are customized to local communities, aiming to improve their effectiveness and applicability in distinct healthcare environments [59]. A study in rural India compared the WHO/ISH laboratory-based and non-laboratory-based CVD risk assessment tools. The results demonstrated a good agreement between the two methods, suggesting that non-laboratory-based risk charts are suitable for CVD risk estimation in resource-limited settings like India [60].

The alternate equation for estimating the WHO/ISH 10-year cardiovascular risk level excludes ASCVD_10 and demonstrates a comparatively slightly reduced predictive power with an $R^2$ of 0.895 and an adjusted $R^2$ of 0.888. Despite this, the equation remains statistically strong, with all predictors showing significant contributions (P=0.000) and low multicollinearity (VIF <1.5). Notably, this equation includes additional variables, such as gender, TC, alongside the previously emphasized factors such as age, smoking status, diabetes status, and systolic blood pressure. As previously reported, the smoking and diabetes status exhibit the highest coefficients (3.177 and 3.983, respectively), confirming their key role in cardiovascular risk. Including gender and TC enhances the model's comprehensiveness, though the slightly lower $R^2$ than the previous model suggests a potential trade-off when ASCVD_10 is omitted. This equation offers an alternative predictive framework, which is particularly useful when ASCVD data is unavailable.

The contribution of specific parameters to the respective risk equations in SEA and SA CVD risk charts revealed notable differences. Age, SBP, R-square, and DW values show similar contributions in both charts. Smoking status and diabetic status exhibit a more pronounced influence in the Southeast Asian chart compared to the South Asian chart. Conversely, TC and HDL cholesterol values contributed to the South Asian risk equations while absent in the Southeast Asian risk prediction equations. This shows the tailored approach of each chart in addressing region-specific risk factors for ASCVD.

Next, a series of ROC curve analyses were performed to establish cut-off values between the hematological and lipid parameters and the risk estimators. A few hematological and biochemical parameters were able to provide cut-off values to predict HDL-LDL ratio for 0.39 cut-off level with significant moderate to high sensitivity, specificity, and acceptable area under curves (AUC >0.600) (**Figure 2** and **Table 2**). In addition, the same cut-off value of the HDL-LDL ratio was used to predict the risk estimator for ASCVD_10 and ASCVD_LT. The ASCVD_10 provided a cut-off value of 4.95 (app. 5.0) with high specificity and acceptable AUC (**Figure 2** and **Table 2**). The cut-off value of ASCVD_10 is in the borderline risk category, while the same LDL falls in the near-optimal range. The ASCVD_LT provided a cut-off value of 37.5 with high sensitivity and acceptable AUC (**Figure 2** and **Table 2**). Further, the ASCVD_LT cut-off value is in the upper borderline of the high-risk category [11]. The ASCVD_OP did not provide reliable results. The use of a 0.39 cut-off value for the HDL-LDL ratio is supported by its predictive power for both ASCVD_10 and the ASCVD_LT. These risk estimators showed cut-off values with high specificity and high sensitivity when using this HDL-LDL ratio cut-off, suggesting that individuals with an HDL-LDL ratio below 0.39 are at a considerably higher risk of developing atherosclerotic cardiovascular disease within the next decade and over their lifetime. This aligns with Das and Ingole (2023) and Panagiotakos and Toutouzas (2003), who empha-

size the importance of HDL-LDL balance in cardiovascular health, as a lower HDL-LDL ratio often indicates a higher proportion of 'bad' cholesterol relative to 'good' cholesterol, thus increasing the risk for cardiovascular events. The obtained cut-off value (0.39) was thought-provoking as it appears just the same as the cut-off value of 0.40 stated in the literature [51]. The results of the ROC curve analysis performed based on the ASCVD_10 risk group categories have shown that the cut-off value of 0.40 (app. 0.39) for HDL-LDL ratio with a high specificity and acceptable AUC. This value has been derived according to the Risk group of ASCVD_10 based on 5.0, in the borderline risk category (**Figure 3**). Interestingly, our results showed that two opposite cut-off values were derived for the HDL-LDL ratio and ASCVD_10, 0.39 and 4.95, with high specificity. Both the values are in the margins of high risk according to the literature [51] and have a reciprocal relationship with each other. These findings convincingly suggest that the easily obtainable HDL LDL ratio could be used to predict the risk levels ASCVD_10. Conversely, using HDL-LDL ratio is beneficial since it is simple, and lipid profile testing is widely accessible in clinical settings.

Since both, the mean values of HDL and LDL were in the upper normal and lower normal ranges, respectively, the behavior of the HDL was tested using the same data in different groups of LDLs from normal to high range (**Table 3**). In both non-parametric Kruskal-Wallis and Mann-Whitney U tests, the analysis showed no significant difference in HDL in any of the aforementioned LDL groups, as shown in **Table 3**. The lack of significant difference in HDL levels across different LDL groups suggests that HDL concentrations might be regulated independently of LDL levels within the low to high ranges we studied. This stability is noteworthy, as HDL is often regarded as 'good' cholesterol due to its role in reverse cholesterol transport and potential protective effects against cardiovascular disease [60, 61]. The findings align with previous literature, which has noted that HDL levels can be influenced by various factors, including genetics, lifestyle, and metabolic conditions, not solely by LDL concentrations [62, 63].

According to **Table 3**, TG levels did not vary significantly across the four LDL groups. This may be due to their distinct regulatory mechanisms and metabolic pathways. Triglycerides are primarily transported in the blood by VLDL produced in the liver. The production and clearance of VLDL particles are influenced by factors such as diet, insulin levels, and liver function. In contrast, LDL cholesterol, which is derived from the conversion of VLDL to LDL, is primarily influenced by the uptake and clearance of LDL particles by LDL receptors in the liver and peripheral tissues [64]. Another reason for the stable TG levels despite variations in LDL could be related to the dietary and metabolic consistency within the study category. If participants had relatively stable dietary habits and physical activity levels, these factors would help maintain consistent TG levels. For instance, diets high in refined carbohydrates and sugars can increase TG levels, while diets rich in healthy fats and regular exercise can help maintain them within a normal range. If these dietary and lifestyle factors did not change significantly among the participants, TG levels would likely remain stable [65]. Equally, genetic factors play a significant role in lipid metabolism, and individuals may have inherent stability in their TG levels due to genetic predispositions, regardless of changes in LDL cholesterol. Certain genetic variants can affect how individuals metabolize fats and produce or clear triglycerides, leading to stable TG levels [66-69]. Additionally, the metabolic processes influencing TG and LDL levels are not always directly correlated. For example, insulin resistance and metabolic syndrome can elevate TG levels independently of LDL [70]. Conversely, factors that specifically affect LDL receptor activity or cholesterol synthesis can alter LDL levels without necessarily impacting TG levels. This independence of metabolic pathways can result in situations where LDL varies while TG remains stable. Our findings are consistent with previous research indicating that triglyceride levels and HDL can remain stable over time, even when other lipid parameters, such as LDL cholesterol, show significant variations. This highlights the complexity of lipid metabolism and the importance of considering multiple lipid parameters when assessing cardiovascular risk [71].

Several limitations could be acknowledged regarding this study. As the study population is limited to a single center with comparatively modest sample size (n=102), it limits the po-

tential of generalization of study findings. This study does not contain a longitudinal follow up to assess the significance of the findings regarding HDL-LDL ratio and future cardiovascular events of the participants. The study did not stratify individuals by genetic conditions, socioeconomic status & dietary patterns, which could be potential confounding factors. To overcome the limitations, future research should increase the sample size and conduct the research as a multi-center study across the country, representing the national population. It would be important in improving the generalizability of the results. The two risk estimators utilized in this study are designed for the individuals who have not met with any CVD previously. By estimating their 10-year risk, there is a strong potential for developing the study as a longitudinal research with follow up for the incidence of any CVDs. The regression derived equation for the WHO 10-year risk, predicted by the Total cholesterol and routinely performed clinical assessments, could be integrated and developed up to electronic mobile health applications. This would lead to scalable, automated CVD risk estimation tool in regional CVD screening programmes, for primary care.

## Conclusions

Strong correlations were observed among the same parameters, i.e., among hematological, biochemical, or risk estimators, not between these parameters. A strong, significant positive correlation between the NLR and PLR and a moderately strong positive correlation between PLT and NLR was observed with increased CVD risk, highlighting these hematological parameters as potential markers for assessing cardiovascular health. ASCVD_10 had a reliable association with two other risk types, ASCVD_OP and ASCVD_LT. Furthermore, we successfully predicted WHO/ISH risk estimators with some of the biochemical, hematological, clinical, biometric, and behavioral risk factors by introducing highly predictive well-fit equations. As previously reported, the smoking pattern and the diabetes status contributed to the output risk values derived from all the equations compared with the other. Finally, the TC-HDL ratio provided a significant correlation with the HDL-LDL ratio that is being studied widely at present to determine CVDs and was able to set a cut-off value for the HDL-LDL ratio to predict the risk levels ASCVD_10 and some of the hematological and biochemical parameters. This finding highlights the potential of the HDL-LDL ratio as a predictive biomarker for identifying individuals at increased risk of ASCVD, enabling early intervention and targeted preventive strategies. In summary, these study findings suggest that specific lipid ratios, hematological and other parameters may provide additional insights into cardiovascular risk prediction. These initial findings should be validated before putting them into practice. This study mainly focused on assessing the ASCVD risk in a low-risk population to identify the primary risk. A follow-up study with a higher number of participants, including high-risk patients, is recommended, and more improved results are expected.

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## Disclosure of conflict of interest

None.

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