BMC Research Notes

RESEARCH NOTE Open Access

Hematological and leukocyte morphometric indices as low-cost diagnostic adjuncts in acute coronary syndromes

Mansi Mehrotra[1], Dushyant Singh Gaur[2], Kunal Gururani[3] and Mansi Kala[4*]

Abstract

Objective Acute Coronary Syndromes (ACS) remain a major cause of morbidity and mortality, particularly in resource-limited settings where access to advanced diagnostics may be constrained. This study aimed to evaluate the utility of routine hematological indices and leukocyte morphometric parameters—volume, conductivity, and scatter (VCS)—as low-cost adjuncts for identifying ACS and differentiating its subtypes.

Results description This cross-sectional study included 291 adults (151 ACS patients and 140 healthy controls) evaluated between January 2022 and August 2023. Compared with controls, ACS patients had significantly higher total leukocyte counts, lower hemoglobin levels, increased mean platelet volume (MPV), reduced neutrophil conductivity (MN-C-NE), and increased monocyte scatter parameters (MN-LALS-MO, MN-AL2-MO) (all $p < 0.01$). On L1-regularized multivariable logistic regression, elevated WBC count, reduced hemoglobin, increased MPV, decreased neutrophil conductivity, and increased monocyte scatter indices independently predicted ACS. The combined model demonstrated excellent discriminatory performance (AUC 0.98 ± 0.02). However, discrimination between ACS subtypes was limited, with poor performance for differentiating STEMI from unstable angina and NSTEMI (AUC 0.63–0.65, $p > 0.05$). These findings suggest that hematological and VCS parameters are effective for identifying ACS but have limited utility in subtype differentiation.

Keywords ACS, STEMI, NSTEMI, Stable angina, VCS, Automated analyser, Morphometry, Leucocytes

Introduction

Acute Coronary Syndromes (ACS) remains the leading cause of morbidity and mortality worldwide, accounting for more than 17 million deaths annually, with the majority occurring in low- and middle-income countries [1]. India, in particular, faces a rapidly escalating ACS burden characterized by earlier disease onset and a higher prevalence among younger populations, posing major challenges for healthcare systems [2]. The pathogenesis of ACS is driven primarily by atherosclerosis, a chronic inflammatory and immune-mediated process involving lipid accumulation, endothelial dysfunction, and leukocyte infiltration of the arterial wall. These processes

*Correspondence:
Mansi Kala
drmansikala@gmail.com
[1]Department of Hematopathology, Homi Bhabha Cancer Hospital, New Chandigarh, Punjab, India
[2]Department of Immunohematology and Blood Transfusion, Himalayan Institute of Medical Sciences, Swami Rama Himalayan University, Swami Ram Nagar, Jolly Grant, Dehradun, Uttarakhand 248016, India
[3]Department of Cardiology, Himalayan Institute of Medical Sciences, Swami Rama Himalayan University, Swami Ram Nagar, Jolly Grant, Dehradun, Uttarakhand 248016, India
[4]Department of Pathology, Himalayan Institute of Medical Sciences, Swami Rama Himalayan University, Swami Ram Nagar, Jolly Grant, Dehradun, Uttarakhand 248016, India

culminate in plaque formation, progression, and eventual rupture, leading to acute thrombotic events such as myocardial infarction (MI) and unstable angina [3]. Vascular smooth muscle cells, inflammatory cytokines, and oxidative stress play central roles in promoting plaque vulnerability [4, 5]. Conventional diagnostic strategies, including electrocardiography, echocardiography, cardiac biomarkers such as troponins, and coronary angiography, remain the cornerstone of ACS evaluation. However, their utility may be limited by cost, accessibility, and sensitivity, especially in early disease detection or prognostication in resource-constrained settings [6]. This has led to increasing interest in identifying inexpensive, readily available biomarkers that reflect systemic inflammation and cardiovascular risk. In this context, hematologic indices—such as the neutrophil-to-lymphocyte ratio (NLR), monocyte-to-lymphocyte ratio (MLR), mean platelet volume (MPV), and red cell distribution width (RDW)—have emerged as potential markers of systemic inflammation, with several studies linking them to ACS severity and adverse outcomes [7–9]. Neutrophils and monocytes, as innate immune effectors, contribute to plaque destabilization, whereas platelet indices reflect thrombogenic potential and endothelial dysfunction [10, 11]. More recently, advanced hematology analyzer–derived parameters, including volume, conductivity, and scatter (VCS) indices, have shown promise in objectively characterizing leukocyte activation and morphology in inflammatory conditions such as sepsis and COVID-19, suggesting potential applicability in cardiovascular disease [12–14].

Given these observations, evaluating hematological parameters and novel VCS-derived indices may offer cost-effective and scalable adjuncts to traditional diagnostics, particularly in resource-limited settings. Because CBC and VCS metrics are already generated during routine testing with no additional consumables or procedures, their use represents minimal incremental cost compared with specialized biomarkers or imaging. The present study aims to assess the role of routine hematological parameters and VCS-derived leukocyte indices in patients with acute coronary syndrome, exploring their ability to differentiate ACS from healthy controls presentations and stratify risk across clinical subgroups.

Materials and methods

This cross-sectional analytical study was conducted between January 2022 and August 2023 in the Departments of Pathology and Cardiology at the Himalayan Institute of Medical Sciences, Swami Rama Himalayan University, and Dehradun, India. Ethical approval was obtained from the Institutional Ethics Committee, and written informed consent was obtained from all participants in accordance with the Declaration of Helsinki.

Study population

A total of 291 adult patients (> 18 years) presenting to the tertiary care center were enrolled. Among these patients, 151 met the diagnostic criteria for ACS, including ST-elevation myocardial infarction (STEMI), non-ST-elevation myocardial infarction (NSTEMI), and unstable angina, which were defined according to the American College of Cardiology (ACC)/American Heart Association (AHA) guidelines [15]. The control group comprised 140 age- and sex-matched healthy volunteers recruited from our hospital's routine health check-up program. All individuals had undergone standardized screening, including clinical history, physical examination, fasting lipid profile, fasting blood glucose, and blood pressure assessment, ensuring exclusion of latent cardiovascular risk factors such as hypertension, diabetes, or dyslipidemia. Additional exclusions included autoimmune diseases, chronic kidney or liver disease, active infection, and any condition known to affect hematological indices.

ACS diagnosis was based on a combination of clinical presentation, ECG findings, and laboratory biomarkers, including cardiac troponin T and creatine phosphokinase-MB (CPK-MB), following internationally accepted criteria [15, 16].

Venous blood samples were collected in EDTA vacutainers and processed within 30 min to minimize preanalytical variability. The samples were analyzed via a Beckman Coulter DxH 800 automated hematology analyzer (Beckman Coulter, California, USA). Routine hematological investigations included hemoglobin (Hb) estimation via spectrophotometry, total leukocyte count (TLC), absolute and differential leukocyte counts, red blood cell indices, and platelet counts. Cell counts and volumes were measured via the Coulter Principle, which determines cell size on the basis of electrical impedance changes as cells suspended in the electrolyte pass through an aperture [17]. Leukocyte volume, conductivity, and scatter (VCS) parameters were obtained using a flow cell and laser-based light-scatter system which assessed neutrophils (NE), lymphocytes (LY), and monocytes (MO). This generated data like mean volume (MN-V), which estimates the average cell size (forward scatter), mean conductivity (MN-C): This metric evaluates the nuclear/cytoplasmic composition via radiofrequency impedance, mean median angle light scatter (MN-MALS), upper median angle (UMALS), lower median angle (LMALS), lower angle (LALS), and axial light loss (AL2). These methods provide quantitative insights into cellular granularity, nuclear segmentation, and cytoplasmic complexity on the basis of multiangle laser scatter analysis. The DxH 800 integrates flow cytometric technology with impedance analysis to obtain VCS-derived leukocyte morphometric data [12, 13]. Internal quality control was

Table 1 Comparison of age distribution between ACS patients and healthy controls

Age range	ACS ($n=151$)	Healthy control ($n=140$)	p – value
31–40	8.62%	5.00%	
41–50	15.79%	15.00%	
51–60	25.82%	48.30%	
61–70	33.77%	25.00%	
> 70	15.99%	6.00%	
Mean age ± SD (years)	58.39 ± 11.39	55.14 ± 8.78	0.08

Table 2 Distribution of key cardiovascular risk factors in ACS patients versus healthy controls

Risk factor	ACS ($n=151$)	Healthy controls ($n=140$)	p-value
Aspirin intake	22 (14.57%)	0 (0%)	< 0.001
Hypertension	40 (26.49%)	0 (0%)	< 0.001
Diabetes mellitus	30 (19.87%)	0 (0%)	< 0.001
Smoking	54 (35.76%)	0 (0%)	< 0.001

performed daily via Coulter Latron controls as per the manufacturer's recommendations [12].

Statistical analysis

The data were compiled via Microsoft Excel and analyzed via SPSS version 22.0. Continuous variables are expressed as the mean ± standard deviation (SD) or median (interquartile range, IQR) depending on normality (tested via the Shapiro–Wilk test). Group comparisons were performed via the independent sample t test or Mann–Whitney U test for continuous variables and the chi-square test for categorical variables. To assess the diagnostic performance of the hematological and VCS parameters, receiver operating characteristic (ROC) curve analysis was performed. The area under the curve (AUC), sensitivity, and specificity were calculated to identify the most discriminative parameters. A p value < 0.05 was considered to indicate statistical significance. For multivariable modeling, penalized logistic regression with L1 regularization (LASSO) was applied to identify independent predictors of ACS status and to evaluate separation between ACS subtypes. Predictors were standardized (z scores), and missing values were imputed using the median. Model performance was assessed via 5-fold stratified cross-validation, and the results are summarized as the means ± SDs of the AUC values. Coefficients from the final fitted models were exponentiated to obtain odds ratios (ORs) per 1-SD increase. A two-sided p value < 0.05 was considered statistically significant.

Results

In the ACS group, the majority of patients (33.77%) were aged 61–70 years, followed by 25.82% in the 51–60-year age bracket. In contrast, the control group had the highest proportion of individuals (48.34%) in the 51–60 years age range. Despite this difference in age distribution, the mean age of ACS patients (58.39 ± 11.39 years) was broadly comparable to that of the control group (55.14 ± 8.78 years). (Table 1). Table 2 explains the key cardiovascular risk factors between ACS patients and healthy controls. A significant difference was observed across all variables. Dyslipidemia status was not consistently recorded for ACS patients in the available clinical data; however, all healthy controls had documented lipid profiles within normal limits as part of the routine health check-up protocol. BMI was not recorded in both the groups. Among the ACS patients, 76.15% were male and 23.84% female. The control group had a slightly different distribution, with 65.71% males and 34.28% females. Most participants, including both ACS patients and healthy controls, were from rural areas (59.31%). Compared with healthy controls, ACS patients had lower hemoglobin levels. Red blood cell (RBC) counts were slightly greater in ACS patients, but hematocrit (HCT) levels were lower than in controls. Compared with controls, ACS patients presented significant differences in MCV, MCHC, RDW, and MPV (Table 3). In contrast to non-ACS controls, patients with ACS demonstrated significantly higher WBC counts, higher absolute neutrophil counts, higher monocyte counts, higher lymphocyte counts and there was a significant difference in the neutrophil, eosinophil and lymphocyte count percentages between the ACS group and the non-ACS group. There was an increase in the mean absolute neutrophil count, absolute lymphocyte count and absolute monocyte count in the ACS patients compared with the healthy control (Table 4). Among the VCS parameters, several showed significant differences between ACS patients and healthy controls. Neutrophil-related indices demonstrated marked changes, including higher MN-V-NE and MN-AL2-NE values in ACS. Similarly, MN-UMALS-NE.

Table 3 Comparison of hematological parameters between ACS patients and healthy controls

	ACS ($n=151$) Mean ± SD	Healthy controls ($n=140$) Mean ± SD	p – value
TLC	9.80 ± 3.86	6.68 ± 1.42	< 0.01
RBC	4.65 ± 1.78	4.53 ± 0.52	0.44
HGB	12.44 ± 2.77	13.80 ± 1.63	< 0.01
HCT	37.40 ± 8.00	41.00 ± 4.31	< 0.01
MCV	87.16 ± 13.50	91.07 ± 9.10	0.004
MCH	32.76 ± 14.01	30.66 ± 3.57	0.08
MCHC	33.15 ± 1.20	33.61 ± 0.91	< 0.01
RDW	16.14 ± 4.90	14.47 ± 1.38	< 0.01
PLT	191.06 ± 80.77	195.68 ± 75.75	0.61
MPV	12.36 ± 1.32	10.89 ± 1.88	< 0.01

TLC: Total Leukocyte Count ($\times 10^9$/L), RBC: Red Blood Cell Count ($\times 10^{12}$/L), HGB: Hemoglobin (g/dl), HCT: Hematocrit (%), MCV: Mean Corpuscular Volume (fl.), MCH: Mean Corpuscular Hemoglobin (pg), MCHC: Mean Corpuscular Hemoglobin Concentration (g/dl), RDW: Red Cell Distribution Width (%), PLT: Platelet Count ($\times 10^3$/L), and MPV: Mean Platelet Volume (fl.)

Table 4 Differential leukocyte parameters in ACS patients compared with healthy controls

	ACS ($n=151$) Mean ± SD	Healthy controls($n=140$) Mean ± SD	p – value
NE%	63.5 ± 17.49	56.70 ± 8.05	0.51
LY%	24.73 ± 14.18	31.57 ± 7.20	0.10
MO%	8.75 ± 4.06	7.57 ± 1.84	< 0.01
EO%	3.36 ± 3.26	3.51 ± 3.07	0.68
BA%	0.74 ± 1.49	0.62 ± 0.29	0.02
NE#	6.35 ± 3.84	3.83 ± 1.15	< 0.01
LY#	2.32 ± 1.86	2.07 ± 0.51	< 0.01
MO#	0.81 ± 0.44	0.50 ± 0.14	< 0.01
EO#	0.24 ± 0.28	0.23 ± 0.22	0.07
BA#	0.07 ± 0.20	0.02 ± 0.04	0.001

NE %: Neutrophils (percentage), LY: Lymphocytes (percentage), MO%: Monocytes (percentage), EO %: Eosinophils (percentage), BA%: Basophils (percentage), NE#: Absolute Neutrophil Count ($\times 10^9$/L), LY#: Absolute Lymphocyte Count ($\times 10^9$/L), MO#: Absolute Monocyte Count ($\times 10^9$/L), EO#: Absolute Eosinophil Count ($\times 10^9$/L), BA#: Absolute Basophil Count ($\times 10^9$/L)

Table 5 Comparison of VCS parameters between ACS and healthy control

VCS parameter (au)	ACS($n=151$) (Mean ± SD)	Healthy control ($n=140$) (Mean ± SD)	p – value
MN-V-NE	152 ± 7.28	149.52 ± 5.23	0.001
MN-C-NE	142.73 ± 3.97	146.07 ± 3.81	< 0.01
MN-MALS-NE	138.87 ± 5.32	140.00 ± 6.91	0.11
MN-UMALS-NE	139.18 ± 5.43	140.95 ± 6.64	0.01
MN-LMALS-NE	134.81 ± 7.09	135.12 ± 8.47	0.71
MN-LALS-NE	164.64 ± 20.39	169.94 ± 30.83	0.07
MN-AL2-NE	142.62 ± 6.91	135.51 ± 7.63	< 0.01
MN-V-LY	90.43 ± 4.40	88.75 ± 3.32	< 0.01
MN-C- LY	114.39 ± 3.89	115.13 ± 4.70	0.14
MN-MALS- LY	71.12 ± 5.53	70.17 ± 7.30	0.22
MN-UMALS- LY	73.6 ± 8.065	73.76 ± 9.10	0.86
MN-LMALS- LY	63.44 ± 4.65	61.17 ± 6.20	< 0.01
MN-AL2- LY	69.42 ± 7.49	65.47 ± 4.90	< 0.01
MN-V-MO	175.44 ± 9.36	171.17 ± 6.28	< 0.01
MN-C- MO	122.14 ± 3.62	123.98 ± 4.01	< 0.01
MN-MALS- MO	89.07 ± 4.56	90.15 ± 5.77	0.07
MN-UMALS- MO	99.09 ± 5.15	99.81 ± 6.66	0.30
MN-LMALS- MO	75.85 ± 5.54	77.07 ± 5.39	0.06
MN-LALS- MO	88.39 ± 15.24	102.32 ± 18.79	< 0.01
MN-AL2- MO	124.78 ± 10.24	119.45 ± 8.25	< 0.01

MN: Mean, V: Volume, C: Conductivity, NE: Neutrophils, LY: Lymphocytes, MO: Monocytes, MALS: Median Angle Light Scatter, UMALS: Upper Median Angle Light Scatter, LMALS: Lower Median Angle Light Scatter, LALS: Low Angle Light Scatter, AL2: Axial Light Loss (secondary axis light scatter measurement

Lymphocyte parameters also showed notable differences, with ACS patients exhibiting higher MN-V-LY and significantly increased MN-LMALS-LY and MN-AL2-LY. ACS patients had significantly higher MN-V-MO and MN-AL2-MO, while MN-C-MO and MN-LALS-MO were significantly lower compared with controls. (Table 5). There was a statistically significant difference in the NLR between the ACS and non-ACS groups (2.57 ± 16.41 and 0.75 ± 1.79, $P < 0.0001$, respectively). There was a statistically significant difference in the lymphocyte monocyte ratio between the ACS and non-ACS groups (3.07 ± 1.8 and 4.96 ± 7.6, $P < 0.0001$, respectively).

In the initial analysis, several hematological and VCS parameters showed statistically significant differences across ACS subgroups. For example, NSTEMI patients demonstrated higher MN-MALS-MO and MN-LMALS-MO values than those with unstable angina, while STEMI patients showed higher WBC counts, RBC counts, MCH, and RDW compared with the unstable angina subgroup. MCV was significantly greater in unstable angina patients compared with STEMI patients, and absolute lymphocyte count was higher in STEMI than in NSTEMI. Additional differences were observed in neutrophil and lymphocyte ratios when each ACS subgroup was compared with healthy controls (Table 6). However, because these comparisons involved a large number of hematological and VCS markers, Bonferroni correction was applied to minimize the risk of false-positive results. After adjustment, most of the initially significant parameters lost statistical significance. The uncorrected p-values are reported for transparency, while the Bonferroni-adjusted values represent the more conservative and statistically robust interpretation of subgroup differences.

To determine the independent contributions of the hematological and leukocyte VCS indices, penalized multiple logistic regression (L1-regularized logistic model) was performed. The model comparing ACS patients (UA, NSTEMI, STEMI) with healthy controls demonstrated excellent discrimination (mean AUC 0.98 ± 0.02,5-fold cross-validation). The key predictors retained in the model were increased total leukocyte count, reduced hemoglobin concentration, elevated mean platelet volume, and neutrophil conductivity and axial light loss parameters, highlighting the central role of inflammation, anemia, platelet activation, and altered leukocyte morphology in ACS pathophysiology (Fig. 1).

When applied to distinguish STEMI from unstable angina, the model achieved only modest predictive performance (mean AUC 0.65 ± 0.07), with monocyte scatter indices and red cell distribution width contributing to the separation. Similarly, the STEMI versus NSTEMI model yielded limited discriminatory ability (mean AUC 0.63 ± 0.07). These findings indicate that while hematological indices and VCS-derived indices are powerful in differentiating ACS patients from healthy control, they are insufficient alone to reliably discriminate among ACS subtypes. Figure 1 shows the ROC curves for the combined models compared with those of selected single predictors (e.g., TLC, MPV, MN-C-NE), demonstrating the incremental diagnostic value of the multivariable approach.

Table 6 Comparative analysis of hematological and VCS parameters across UA, NSTEMI, and STEMI groups

Parameter	UA vs NSTEMI (p)	UA vs STEMI (p)	STEMI vs NSTEMI (p)	UA vs NSTEMI (Bonferroni)	UA vs STEMI (Bonferroni)	STEMI vs NSTEMI (Bonferroni)
TLC	0.09	0.001	0.216	1	0.123	1
RBC	0.532	0.045	0.076	1	1	1
HGB	0.741	0.064	0.083	1	1	1
HCT	0.789	0.077	0.1	1	1	1
MCV	0.164	0.03	0.012	1	1	1
MCH	0.211	0.033	0.153	1	1	1
MCHC	0.958	0.07	0.119	1	1	1
RDW	0.475	0.011	0.108	1	1	1
PLT	0.588	0.055	0.365	1	1	1
MPV	0.23	0.407	0.618	1	1	1
NE	0.356	0.221	0.118	1	1	1
LY	0.42	0.116	0.08	1	1	1
MO	0.931	0.885	0.856	1	1	1
EO	0.214	0.771	0.155	1	1	1
BA	0.339	0.085	0.13	1	1	1
NE#	0.073	0.091	0.886	1	1	1
LY#	0.621	0.01	0.023	1	1	1
MO#	0.725	0.036	0.142	1	1	1
EO#	0.184	0.862	0.119	1	1	1
BA#	0.921	0.055	0.125	1	1	1
MN-V-NE	0.308	0.373	0.689	1	1	1
MN-C-NE	0.365	0.896	0.303	1	1	1
MN-MALS-NE	0.661	0.672	0.435	1	1	1
MN-UMALS-NE	0.435	0.846	0.559	1	1	1
MN-LMALS-NE	0.827	0.473	0.441	1	1	1
MN-LALS-NE	0.267	0.62	0.48	1	1	1
MN-AL2-NE	0.537	0.473	0.262	1	1	1
MN-V-LY	0.455	0.568	0.81	1	1	1
MN-C-LY	0.169	0.191	0.731	1	1	1
MN-MALS-LY	0.298	0.267	0.932	1	1	1
MN-UMALS-LY	0.408	0.322	0.983	1	1	1
MN-LMALS-LY	0.12	0.319	0.504	1	1	1
MN-LALS-LY	0.277	0.578	0.45	1	1	1
MN-AL2-LY	0.912	0.332	0.367	1	1	1
MN-V-MO	0.414	0.847	0.517	1	1	1
MN-C-MO	0.666	0.894	0.584	1	1	1
MN-MALS-MO	0.006	0.487	0.061	0.738	1	1
MN-UMALS-MO	0.078	0.485	0.289	1	1	1
MN-LMALS-MO	0.003	0.77	0.015	0.369	1	1
MN-LALS-MO	0.142	0.582	0.042	1	1	1
MN-AL2-MO	0.427	0.286	0.13	1	1	1

Discussion

Our study highlights significant hematological and morphometric differences between patients with ACS and healthy controls, with additional insights from leukocyte volume, conductivity, and scatter (VCS) indices. ACS patients presented higher white blood cell (WBC) counts; increased absolute neutrophil, lymphocyte, and monocyte counts; lower hemoglobin; and elevated mean platelet volume (MPV) and red cell distribution width (RDW) than controls did, reinforcing the role of systemic inflammation in ACS pathophysiology [1–5].

Neutrophils, the predominant WBC subset and principal effectors in acute inflammation, secrete cytokines and enzymes that increase leukocyte recruitment and promote atherosclerotic plaque destabilization [10, 18]. Historical and contemporary studies consistently demonstrate that higher leukocyte counts predict myocardial infarction and adverse cardiovascular outcomes [19–21]. Our findings corroborate these associations, with elevated neutrophil counts and higher NLRs in ACS patients than in controls. However, no significant differences in neutrophil VCS parameters (mean volume, conductivity, scatter) were observed among ACS subtypes (STEMI, NSTEMI, unstable angina), suggesting that while inflammation is a hallmark of ACS, VCS-derived neutrophil morphology alone may not discriminate ACS phenotypes, a result that is not fully consistent with prior reports of phenotypic neutrophil variation in ACS [22].

Monocytes, which are central to atherogenesis, exhibited elevated absolute counts in ACS patients. Our VCS analysis revealed increased mean volume and altered scatter indices of monocytes in ACS patients compared with controls. Importantly, NSTEMI patients demonstrated significantly greater monocyte scatter (MN-MALS, LMALS) than unstable angina and STEMI patients did, suggesting subtype-specific differences in monocyte activation or nuclear complexity. Prior studies have emphasized the pathogenic role of intermediate and non-classical monocytes in promoting plaque instability and adverse outcomes [23, 24]; Several hematological and VCS parameters differed across ACS subgroups in our study, but these findings must be interpreted cautiously due to potential confounders such as inflammation, comorbidities, medication use, and pre-analytical variability.

Even so, the biological plausibility remains strong, as neutrophil and monocyte activation, platelet reactivity, and stress-related lymphocyte changes are well-recognized in ACS and align with the patterns observed in this study. Notably, although some parameters were initially significant, most lost significance after Bonferroni correction, indicating that the uncorrected results

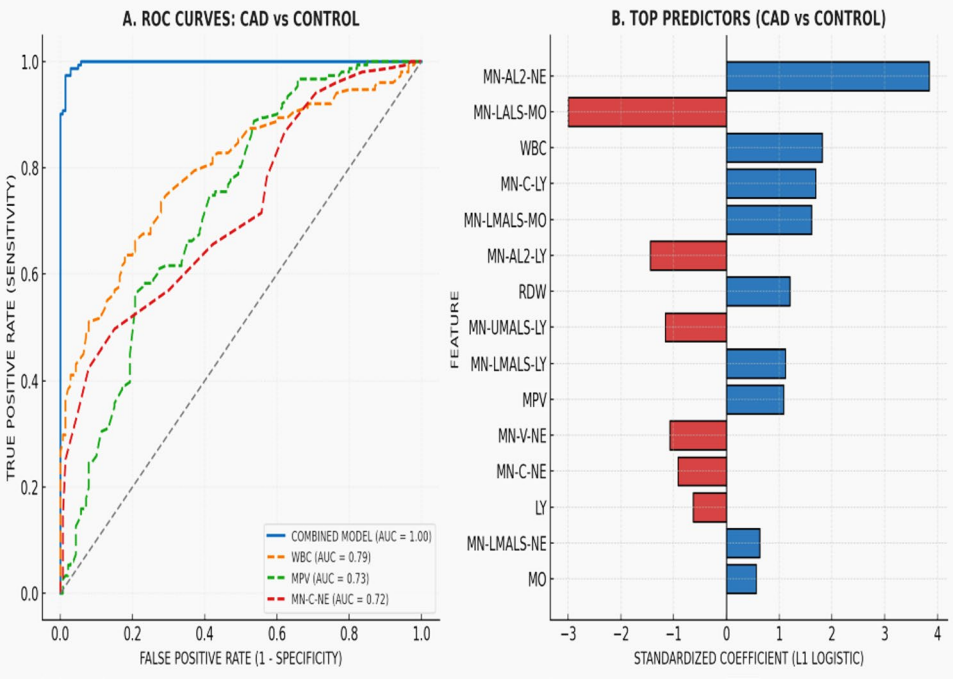

Fig. 1 ROC Curves and Predictor Importance for ACS vs. Healthy Control Classification. **A** Receiver Operating Characteristic (ROC) curves comparing the performance of individual hematological/VCS parameters and their combination in differentiating ACS from healthy control. The combined model achieved an AUC of 1.00, while individual predictors such as WBC (AUC = 0.79), MPV (AUC = 0.72), and MN-C-NE (AUC = 0.72) showed moderate discriminative ability. **B** Standardized coefficients (L1 logistic regression) of the top predictors in the combined model. MN-AL2-NE, MN-LALS-MO, and MN-C-LY emerged as the strongest positive predictors, whereas features such as MN-UMALS-MO and WBC contributed negatively to classification

may reflect multiple testing effects. The corrected values therefore provide a more conservative interpretation, and larger studies with external validation are needed to confirm these associations.

Both the NLR and MLR were elevated in ACS patients compared with controls, which is consistent with the literature linking these ratios to cardiovascular risk and adverse outcomes following ACS and PCI [25–28]. Although our study did not establish predictive cutoffs for the NLR or MLR within subgroups, these findings reinforce their utility as accessible inflammatory biomarkers in ACS management.

The increased RDW and MPV in ACS patients align with evidence that anisocytosis and platelet activation reflect underlying endothelial dysfunction and thrombotic propensity [29, 30]. Interestingly, we observed slightly higher RBC counts in ACS patients than in studies reporting lower RBC counts in patients with advanced disease [4]. This discrepancy may reflect population differences or early disease representations in our cohort.

Our study is the first to systematically analyze leukocyte VCS parameters in ACS and ACS patients. While these parameters have been evaluated in sepsis, COVID-19, and other infectious conditions [12–14], their role in cardiovascular disease remains underexplored. We observed greater mean neutrophil and monocyte volumes and altered conductivity in ACS patients, paralleling the inflammatory signatures described in sepsis and COVID-19 patients [13, 14]. This convergence suggests that leukocyte morphometric remodeling is a nonspecific but conserved feature of systemic inflammation. However, unlike in sepsis, where neutrophil VCS cutoffs achieve strong diagnostic utility [12].

In the multivariable L1-regularized logistic regression model, elevated WBC count, reduced hemoglobin, higher MPV, and lower neutrophil conductivity (MN-C-NE) emerged as independent predictors of ACS, along with monocyte and neutrophil scatter indices such as MN-LALS-MO and MN-AL2-NE respectively. These associations reflect biologically plausible mechanisms involving leukocyte activation, anemia, platelet reactivity, and inflammatory cell morphodynamics. However, when the same approach was applied to distinguish between ACS subtypes, no predictors retained statistical significance (all $p > 0.05$), consistent with the limited discriminatory performance of these models (AUC 0.63–0.65). Although the combined model for CAD vs. non-CAD initially showed a very high AUC, this likely reflects optimistic bias inherent in high-dimensional data and emphasizes the need for cautious interpretation and external validation. Importantly, routine hematological indices and VCS parameters remain inexpensive, rapidly obtainable, and widely available on standard hematology analyzers, supporting their potential role as low-cost

adjunct diagnostic tools in settings where advanced biomarkers or imaging may be limited. Future studies with larger cohorts and fully nested validation frameworks should further evaluate specific VCS signatures, explore integration with machine-learning algorithms, and determine their prognostic value in long-term cardiovascular risk stratification.

Conclusion

In summary, hematological indices and leukocyte VCS parameters demonstrated meaningful discriminatory ability for identifying ACS, with several markers—including elevated WBC counts, reduced hemoglobin, higher MPV, and altered neutrophil and monocyte scatter indices—emerging as independent predictors. These findings reflect the central role of inflammation, anemia, platelet activation, and leukocyte morphologic changes in ACS. Although the multivariable model showed high apparent accuracy, this performance is likely influenced by optimistic bias inherent in high-dimensional data and therefore requires cautious interpretation and external validation. In contrast, the ability of these parameters to differentiate ACS subtypes was modest, indicating that they are best used in conjunction with clinical, ECG, and biochemical information for effective risk stratification. Given their low cost and widespread availability on routine hematology analyzers, hematological and VCS metrics hold promise as accessible adjunct tools for early ACS assessment, particularly in resource-limited settings. Larger, prospective, multicenter studies with rigorous validation frameworks are needed to confirm their diagnostic and prognostic utility.

Limitations of the study

This study has several limitations. First, the sample size—particularly within ACS subgroups—was modest, which may have reduced the power to detect meaningful differences between STEMI, NSTEMI, and unstable angina. Second, although median imputation was performed to address missing clinical variables, the high proportion of missingness for comorbidities and medication history. Furthermore, dyslipidemia status was incompletely documented among ACS patients, and BMI values were unavailable for both groups. The absence of these key metabolic risk variables restricts comprehensive adjustment for confounding and may influence the observed associations. Third, the multivariable model demonstrated a very high apparent AUC, raising the possibility of optimistic bias or subtle data leakage, as preprocessing steps may not have been fully nested within the cross-validation procedure. Fourth, many initially significant associations did not remain significant after Bonferroni correction, indicating that several findings should be interpreted as exploratory. Fifth, VCS parameters may be influenced by pre-analytical factors such as sample handling time and underlying inflammatory states, which were not fully controlled in this retrospective dataset. Finally, the absence of external validation limits the generalizability of the predictive model and the VCS signatures identified. Larger, prospective, multicenter studies with rigorous validation frameworks are needed to verify these finding.

Acknowledgements

The authors thank the staff and patients of the Himalayan Institute of Medical Sciences for their support.

Author contributions

MM conceptualized and designed the study, supervised laboratory work and data collection, performed data analysis, and drafted the initial manuscript.DSG contributed to study design, provided expert guidance in immunohematology and transfusion medicine, supervised laboratory interpretation, and critically revised the manuscript for important intellectual content.KG conceptualized the study, contributed to study design, provided cardiology expertise, assisted in clinical interpretation of findings, and reviewed and edited the manuscript.MK conceptualized the study, contributed to study design and methodology, provided senior academic oversight, supervised pathological interpretation, and critically revised the manuscript for intellectual content.All authors read and approved the final manuscript and agree to be accountable for all aspects of the work.

Funding

No funding was received for this study.

Data availability

The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

Declarations

Ethics approval and consent to participate

Ethical approval for the study was obtained from the Institutional Ethics Committee of Himalayan Institute of Medical Sciences, Swami Rama Himalayan University (Approval No. SRHU/HIMS/RC/ETHICS/2022/297). All procedures involving human participants were conducted in accordance with the ethical standards of the institutional research committee and with the 1964 Declaration of Helsinki and its later amendments or comparable ethical standards. The research committee reviewed and approved the study on 15 September 2022. Written informed consent was obtained from all patients and healthy controls prior to participation, including for peripheral blood examination, in accordance with institutional guidelines. Participation was voluntary, and confidentiality was ensured through anonymization of data and secure handling of study records.

Informed consent

As per our policy an informed consent is obtained from all the patients before any invasive procedures such as bone marrow biopsy and or blood transfusions.

Consent for publication

Written informed consent was obtained from all patients and healthy controls prior to publication including for peripheral blood examination, in accordance with institutional guidelines. Participation was voluntary, and confidentiality was ensured through anonymization of data and secure handling of study records.

Competing interests

The authors declare no competing interests.

Received: 22 August 2025 / Accepted: 5 February 2026
Published online: 02 May 2026

References

1. Roth GA, Mensah GA, Johnson CO, et al. Global burden of cardiovascular diseases and risk factors, 1990–2022. J Am Coll Cardiol. 2024;83(2):215–39. https://doi.org/10.1016/j.jacc.2023.10.001.
2. Prabhakaran D, Jeemon P, Roy A. Cardiovascular diseases in india: current epidemiology and future directions. Circulation. 2022;145(19):1510–2. https://doi.org/10.1161/CIRCULATIONAHA.121.057289.
3. Libby P, Buring JE, Badimon L, et al. Atherosclerosis Nat Rev Dis Primers. 2019;5(1):56. https://doi.org/10.1038/s41572-019-0106-z.
4. Doran AC, Meller N, McNamara CA. Role of smooth muscle cells in the initiation and early progression of atherosclerosis. Arterioscler Thromb Vasc Biol. 2008;28(5):812–9. https://doi.org/10.1161/ATVBAHA.107.159327.
5. Ridker PM, Inflammation. C-reactive protein, and cardiovascular risk: moving toward clarity. N Engl J Med. 2023;389(1):29–40. https://doi.org/10.1056/NEJMra2301383.
6. Alqahtani A, Bakr M, Ibrahim A, et al. Diagnostic performance of cardiac troponin and ECG in acute coronary syndrome: a meta-analysis. Heart Lung. 2023;60:105260. https://doi.org/10.1016/j.hrtlng.2023.105260.
7. Zhao Y, Xiang Y, Pan Z, et al. Hematologic indices as prognostic markers in coronary artery disease: A systematic review and meta-analysis. Am J Cardiol. 2023;197:1–9. https://doi.org/10.1016/j.amjcard.2023.01.030.
8. Wang QC, Wang ZY. Comparative analysis of neutrophil-to-lymphocyte ratio and remnant cholesterol in predicting cardiovascular events and mortality. Sci Rep. 2023;13:22362. https://doi.org/10.1038/s41598-023-49403-8.
9. Fan Z, Ji H, Ma S, et al. Monocyte-to-lymphocyte ratio and neutrophil-to-lymphocyte ratio in NSTEMI after PCI. BMJ Open. 2018;8:e023459. https://doi.org/10.1136/bmjopen-2018-023459.
10. Rosales C. Neutrophil: a cell with many roles in inflammation. Front Physiol. 2018;9:113. https://doi.org/10.3389/fphys.2018.00113.
11. Pafili K, Penlioglou T, Papanas N. Mean platelet volume and coronary artery disease. CurrOpinCardiol. 2019;34(4):390–8. https://doi.org/10.1097/HCO.0000000000000624.
12. Lippi G, Plebani M. Volume, conductivity, and scatter parameters of leukocytes in clinical diagnostics. Clin Chem Lab Med. 2020;58(2):182–91. https://doi.org/10.1515/cclm-2019-0661.
13. Mathur A, Thakur M, Raina V, et al. Leucocyte volume, conductivity, and scatter at admission predicts outcome in COVID-19 patients: a prospective study. Niger J Clin Pract. 2023;26(6):857–63. https://doi.org/10.4103/njcp.njcp_2268_22.
14. Ranjan P, Kumari S, Sahu SK, et al. Utility of volume, conductivity, and scatter parameters in sepsis diagnosis: a systematic review. Front Med (Lausanne). 2022;9:935641. https://doi.org/10.3389/fmed.2022.935641.
15. Amsterdam EA, Wenger NK, Brindis RG, et al. 2014 AHA/ACC guideline for the management of patients with non–ST-elevation acute coronary syndromes. J Am Coll Cardiol. 2014;64(24):e139–228. https://doi.org/10.1016/j.jacc.2014.09.017.
16. Thygesen K, Alpert JS, Jaffe AS, et al. Fourth universal definition of myocardial infarction (2018). Eur Heart J. 2019;40(3):237–69. https://doi.org/10.1093/eurheartj/ehy462.
17. Brown C, Hollingsworth J. Coulter principle for cell counting and sizing. J Lab Med. 2019;43(2):73–82. https://doi.org/10.1515/labmed-2019-0105.
18. Soehnlein O. Multiple roles for neutrophils in atherosclerosis. Circ Res. 2012;110(6):875–88. https://doi.org/10.1161/CIRCRESAHA.111.257535.
19. Friedman GD, Klatsky AL, Siegelaub AB. The leukocyte count as a predictor of myocardial infarction. N Engl J Med. 1974;290:1275–8. https://doi.org/10.1056/NEJM197406062902302.
20. Luo J, Thomassen JQ, Nordestgaard BG, et al. Neutrophil counts and cardiovascular disease. Eur Heart J. 2023;44(47):4953–64. https://doi.org/10.1093/eurheartj/ehad649.
21. Welsh C, Welsh P, Mark PB, et al. Association of total and differential leukocyte counts with cardiovascular disease and mortality in the UK biobank. Arterioscler Thromb Vasc Biol. 2018;38(6):1415–23. https://doi.org/10.1161/ATVBAHA.118.310945.
22. Maréchal P, Tridetti J, Nguyen ML, et al. Neutrophil phenotypes in coronary artery disease. J Clin Med. 2020;9(5):1602. https://doi.org/10.3390/jcm9051602.
23. Amengual J, Barrett TJ. Monocytes and macrophages in atherogenesis. Curr Opin Lipidol. 2019;30(5):401–8. https://doi.org/10.1097/MOL.0000000000000634.
24. Idzkowska E, Eljaszewicz A, Miklasz P, et al. The role of different monocyte subsets in the pathogenesis of atherosclerosis and acute coronary syndromes. Scand J Immunol. 2015;82(3):163–73. https://doi.org/10.1111/sji.12314.
25. Bhat T, Teli S, Rijal J, et al. Neutrophil to lymphocyte ratio and cardiovascular diseases: a review. Expert Rev Cardiovasc Ther. 2013;11(1):55–9. https://doi.org/10.1586/erc.12.159.
26. Buonacera A, Stancanelli B, Colaci M, Malatino L. Neutrophil-to-lymphocyte ratio: an emerging marker of immune system–disease interactions. Int J Mol Sci. 2022;23(7):3636. https://doi.org/10.3390/ijms23073636.
27. Chen H, Li M, Liu L, et al. Monocyte/lymphocyte ratio and severity of coronary artery disease in NSTEMI. Med (Baltim). 2019;98(26):e16267. https://doi.org/10.1097/MD.0000000000016267.
28. Gao X, Liu Y, Tian Y, et al. Prognostic value of peripheral blood inflammatory cell subsets in ACS after PCI. J Int Med Res. 2021;49(4):3000605211010059. https://doi.org/10.1177/03000605211010059.
29. Dziedzic EA, Gąsior JS, Tuzimek A, Kochman W. Blood count–derived inflammatory markers and acute complications of ischemic heart disease in elderly women. J Clin Med. 2023;12(4):1369. https://doi.org/10.3390/jcm12041369.
30. Sincer I, Gunes Y, Mansiroglu AK, et al. Association of mean platelet volume and red blood cell distribution width with coronary collateral development in stable coronary artery disease. Postepy Kardiol Interwencyjnej. 2018;14(3):263–9. https://doi.org/10.5114/aic.2018.78329.

Publisher's Note