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Systemic Laboratory Alterations Associated With Heart Failure in Severe Mitral Regurgitation: A Retrospective Observational Study

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Emilia Sorina Fiat [1, 2], Nastaca Alina Palade [3], Crina Cristina Solomon [3], Razvan Constantin Vonica [1], Bogdan Ioan Vintila [4], Maria Totan [5], Anca Maria Arseniu [1], Claudiu Morgovan [6], Anca Butuca [6], Adina Frum [1], Carmen Maximiliana Dobrea [1], Diter Atasie [7, 1], Felicia Gabriela Gligor [6]

1. Faculty of Medicine, Lucian Blaga University of Sibiu, Sibiu, ROU 2. Cardiology, County Clinical Emergency Hospital, Sibiu, ROU 3. Preclinical Medicine, Lucian Blaga University of Sibiu, Sibiu, ROU 4. Anesthesia and Critical Care, Emergency County Clinical University of Sibiu, Sibiu, ROU 5. Biochemistry, Faculty of Medicine, Lucian Blaga University of Sibiu, Sibiu, ROU 6. Preclinical Medicine, Faculty of Medicine, Lucian Blaga University of Sibiu, Sibiu, ROU 7. Medical Genetics, MedLife Polisano Hospital, Sibiu, ROU

Corresponding author: Nastaca Alina Palade, nastacaalina.coman@ulbsibiu.ro

Abstract

Background: The systemic laboratory abnormalities accompanying documented heart failure in patients hospitalized with severe mitral regurgitation (MR) have not been fully characterized across multiple biological domains within the same cohort.

Methodology: In this retrospective, single-center, cross-sectional study, 107 adults with echocardiographically documented severe MR were compared according to the presence (n = 49) or absence (n = 58) of a heart failure diagnosis at discharge. Inflammatory, hematological, renal, electrolyte, hepatic, and coagulation measures were evaluated using multiplicity-adjusted comparisons and age- and sex-adjusted domain-specific logistic regression. A post hoc model integrating age, sex, C-reactive protein, anemia, and estimated glomerular filtration rate (eGFR) <60 mL/min/1.73 m² was explored using sensitivity analyses and fixed-model bootstrap validation.

Results: Multiple laboratory domains differed by heart failure status. In the post hoc integrated model, eGFR <escape><</escape>60 mL/min/1.73 m² showed the most consistent association with concomitant heart failure (odds ratio, 5.56; 95% confidence interval, 1.94-15.94; P = 0.001); C-reactive protein and anemia were not statistically significant after mutual adjustment. The apparent and optimism-corrected areas under the curve were 0.809 and 0.780, respectively; corresponding Brier scores were 0.175 and 0.196.

Conclusions: Among hospitalized patients with severe MR, documented concomitant heart failure was accompanied by abnormalities across several laboratory domains, with reduced renal function showing the most consistent association in exploratory multivariable analyses. The retrospective design, discharge-diagnosis-based heart failure classification, incomplete echocardiographic characterization, and limited sample size preclude causal or predictive interpretations.

Categories: Cardiology, Internal Medicine, Nephrology
Keywords: anemia, biomarkers, congestive heart failure, estimated glomerular filtration rate, mitral regurgitation, neutrophil-to-lymphocyte ratio

Introduction

Mitral regurgitation (MR) is one of the most common valvular heart diseases and an important cause of cardiovascular morbidity worldwide. MR occurs when blood flows backward from the left ventricle into the left atrium during systole. Although mild MR is frequently detected and may remain clinically stable, moderate-to-severe MR is associated with progressive cardiac remodeling, reduced functional capacity, heart failure, and increased mortality. In Europe, a systematic review and meta-analysis of population-based studies estimated the crude prevalence of moderate-to-severe MR at 0.60% (95% confidence interval (CI), 0.34-0.92), lower than in North America (1.11%) but rising steeply with age, reaching approximately 2.85% at age 70 and 6.45% by age 90 [1]. The clinical course of severe MR is highly heterogeneous, with some patients remaining stable for years whereas others present with concomitant congestive heart failure (CHF), which is associated with considerable morbidity and mortality. Clinical and echocardiographic assessment remains central to the evaluation of severe MR; however, routinely available laboratory parameters may provide additional information regarding the systemic abnormalities accompanying CHF in this population [2-5].

In severe MR, chronic volume overload of the left atrium and left ventricle promotes progressive cardiac remodeling and, when heart failure develops, leads to elevated left atrial and pulmonary venous pressures,

reduced forward stroke volume, and eventually systemic venous congestion. These hemodynamic consequences provide a biological basis for abnormalities across several laboratory domains. Reduced renal perfusion and renal venous congestion may impair kidney function, and neurohormonal activation may promote hyponatremia and other electrolyte disturbances. Hepatic congestion or hypoperfusion may alter aminotransferase levels and the hepatic synthesis of coagulation factors. Systemic inflammation may affect leukocyte subsets, acute-phase reactants, and erythropoiesis. Because these processes may occur simultaneously and influence one another, examining a single laboratory domain may provide only a partial description of the systemic involvement accompanying CHF in this population. Comorbidities and treatment exposures may also influence the observed profiles [2,5-10].

Routine laboratory biomarkers may provide insight into these systemic processes. Acute-phase reactants such as C-reactive protein (CRP) and fibrinogen reflect systemic inflammatory activation and have been associated with adverse cardiovascular outcomes. CRP is synthesized in the liver in response to interleukin-6 (IL-6) and is widely used as a nonspecific marker of systemic inflammation. Fibrinogen promotes a state of hypercoagulability, increasing plasma viscosity and platelet aggregation. High baseline levels of fibrinogen have been established as an independent risk factor for cardiovascular events and mortality [11-14]. Because these biomarkers are not specific to heart failure, their interpretation requires consideration of comorbidities, treatment exposure, and the broader clinical context.

Complete blood count (CBC)-derived inflammatory indices, including the neutrophil-to-lymphocyte ratio (NLR), platelet-to-lymphocyte ratio (PLR), monocyte-to-lymphocyte ratio (MLR), systemic immune-inflammation index (SII), and systemic inflammation response index (SIRI), can be calculated from routine measurements, although indices sharing the same cellular components provide overlapping information. Beyond inflammation, anemia and increased red cell distribution width may reflect inflammation, altered erythropoiesis, or renal impairment, whereas renal, electrolyte, hepatic, and coagulation abnormalities may reflect reduced perfusion, systemic congestion, disease severity, comorbidities, or treatment exposure [15-21].

Several circulating biomarkers have already been studied in heart failure and MR. Natriuretic peptides, particularly B-type natriuretic peptide (BNP) and N-terminal pro-B-type natriuretic peptide (NT-proBNP), are established adjuncts for heart failure assessment and risk stratification, including in valvular heart disease [22]. In the BIOSTAT-CHF cohorts, patients with heart failure and moderate-to-severe MR had higher levels of NT-proBNP, CA125, fibroblast growth factor 23, and growth hormone 1 and lower levels of leptin than those with no or mild MR, reflecting differences in congestion, lipid and mineral metabolism, and oxidative stress [23]; the prognostic impact of MR in worsening heart failure has also been examined in this program [4]. These studies compared patients with heart failure according to the presence of MR, whereas the present study compared patients who all had severe MR according to concomitant CHF status. These investigations provide relevant context but do not directly answer the present cross-sectional question of how routine laboratory abnormalities across several domains differ between hospitalized patients who all have severe MR according to concomitant CHF status. BNP and NT-proBNP were not available for systematic analysis in our cohort; accordingly, the present findings should not be interpreted as a comparison with or alternative to established cardiac biomarkers.

Although laboratory abnormalities have been extensively described in general heart failure populations, their joint characterization specifically among patients with severe MR, stratified by concomitant CHF status, remains insufficiently established. The scientific rationale for a multidomain assessment is that inflammatory activation, altered hematological indices, renal dysfunction, electrolyte changes, hepatic abnormalities, and coagulation disturbances may coexist in this setting and may provide overlapping, nonspecific information about systemic involvement. We therefore compared routinely available parameters across these domains in hospitalized patients with severe MR, grouped according to the presence or absence of a documented CHF discharge diagnosis. The primary objective was to characterize cross-sectional between-group differences and selected age- and sex-adjusted associations. Secondary exploratory objectives were to examine correlations among biomarkers, assess associations in a post hoc integrated model, and evaluate their robustness through sensitivity analyses and fixed-model bootstrap validation. The study was not designed to establish causality, incremental clinical value over echocardiography, or prospective prediction of heart failure beyond conventional cardiac assessment.

Materials And Methods

Study design and population

This retrospective, single-center, cross-sectional study was conducted in the Department of Cardiology of Sibiu County Clinical Emergency Hospital, Romania. Adult patients hospitalized between 21 January and 31 December 2025 who were identified in the hospital information system as having a diagnosis of MR, either coded as a principal or secondary diagnosis or documented in the medical record, were consecutively screened for eligibility (n = 429). Of these, 118 were excluded because MR was not the principal diagnosis of the admission, leaving 311 patients with a principal diagnosis of MR. Among them, 204 were excluded because severe MR was not confirmed, and 107 patients with severe MR were included in the final analysis cohort. Consequently, patients admitted with heart failure as the principal diagnosis and concomitant

severe secondary MR were not included. Of the included patients, 49 had documented concomitant CHF, whereas 58 did not. The patient-selection process and reasons for exclusion are presented in Figure 1.

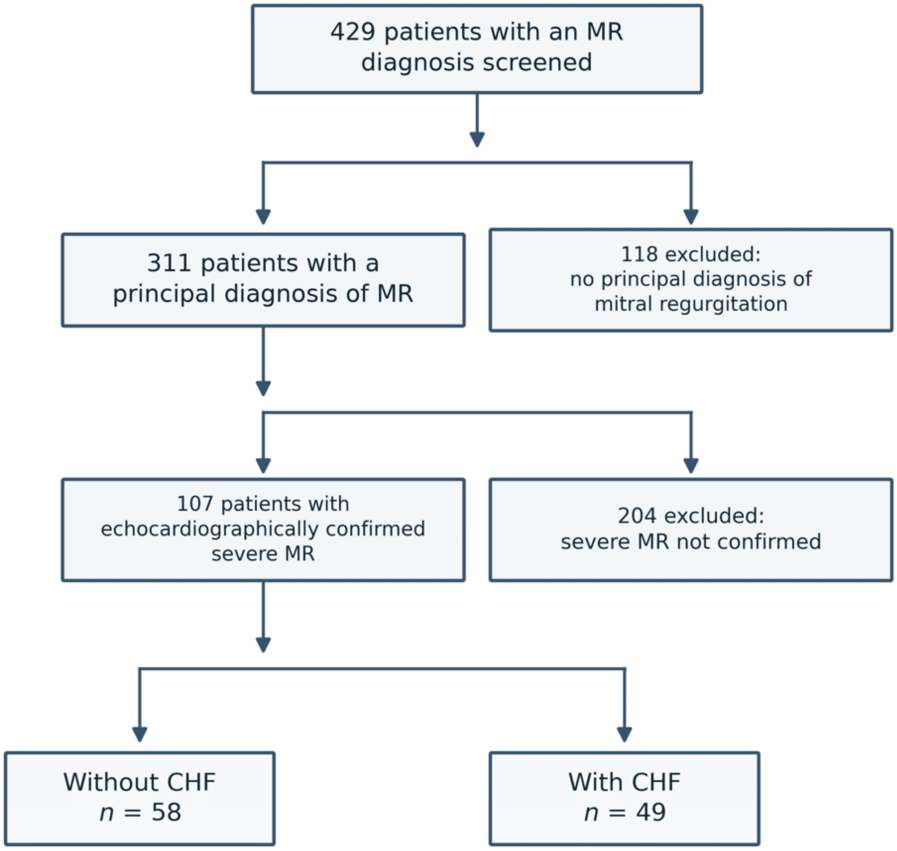


FIGURE 1: Flow diagram of patient screening, eligibility assessment, and inclusion in the final study cohort.

MR, mitral regurgitation; CHF, congestive heart failure; *n*, number of patients

Eligible patients met all inclusion criteria and none of the exclusion criteria, which are summarized in Table 1. Patient identifiers were reviewed within the final cohort, and none of the 107 included patients had more than one hospitalization during the study period. Each patient therefore contributed one unique hospitalization to the analysis, hereafter referred to as the index hospitalization.

Criterion type	Criterion
Inclusion	Age ≥18 years
Inclusion	Hospitalization in the Department of Cardiology of Sibiu County Clinical Emergency Hospital between January 21 and December 31, 2025
Inclusion	Severe mitral regurgitation documented by transthoracic echocardiography during the index hospitalization
Exclusion	No principal diagnosis of mitral regurgitation during the hospitalization
Exclusion	Mitral regurgitation not confirmed as severe by transthoracic echocardiography during the hospitalization

TABLE 1: Inclusion and exclusion criteria of the study.

The index hospitalization was the single hospitalization contributed by each patient during the study period.

All 107 patients included in the final cohort had complete demographic and laboratory data for the variables used in the comparative and multivariable analyses. Accordingly, no imputation of missing laboratory values was performed.

Ascertainment of congestive heart failure

CHF status was retrospectively ascertained from the final diagnoses recorded in the electronic medical records at hospital discharge. The diagnosis was established by the treating cardiology team during the index hospitalization based on the overall clinical assessment and investigations performed as part of routine clinical care.

Patients with a documented discharge diagnosis of CHF were assigned to the CHF group, whereas patients without such a diagnosis were assigned to the non-CHF group. CHF status was not independently adjudicated according to a prespecified research definition.

The available retrospective records did not consistently distinguish acute or decompensated CHF from chronic or clinically stable CHF. Heart failure phenotype according to left ventricular ejection fraction and New York Heart Association (NYHA) functional class [24] was also not systematically available and was therefore not incorporated into the group definition or subgroup analyses. CHF was consequently treated as a contemporaneously documented clinical status rather than as a prospectively adjudicated outcome.

Echocardiographic assessment of severe mitral regurgitation

Severe MR on transthoracic echocardiography performed during the index hospitalization was recorded in the medical records of all included patients. The full echocardiographic report was retrievable for 51 of the 107 patients; in the remaining 56 patients, severe MR was confirmed from the echocardiographic diagnosis documented in the discharge summary, without access to the complete report. Echocardiographic examinations were performed and interpreted by cardiologists from the Department of Cardiology as part of routine clinical care.

MR severity was determined from the final clinical echocardiographic assessment, which integrated valve morphology, color Doppler findings, and the quantitative or semiquantitative parameters available for the individual patient. Because the source reports were unstructured free-text clinical documents, the availability of individual measurements varied across patients, and the complete quantitative criteria used for the original clinical classification could not be retrospectively verified in every case.

Free-text echocardiographic reports were retrievable for 51 of the 107 patients (47.7%), and left ventricular ejection fraction could be extracted for 47 patients (43.9%). Quantitative MR measurements, including effective regurgitant orifice area, regurgitant volume determined using the proximal isovelocity surface area method, vena contracta width, and mitral annulus dimensions, were not consistently available. Left ventricular and left atrial volumes, pulmonary artery pressure, and indices of right ventricular systolic function were similarly unavailable for systematic extraction.

MR etiology, including classification as primary or secondary and as ischemic or non-ischemic, could not be established consistently from the available records. Therefore, these variables were not included in the comparative or multivariable analyses.

Transesophageal echocardiography was additionally performed in 6 of 107 patients (5.6%), generally to further characterize the mechanism of MR or guide intervention planning. For most patients, the classification of severe MR was based on transthoracic echocardiography performed during routine clinical care.

Data collection and laboratory assessment

The analysis was based on an anonymized dataset compiled retrospectively from the patients' hospital medical records. The dataset contained demographic characteristics, documented diagnoses, laboratory values, echocardiographic information, comorbidities, and pharmacological treatments associated with the index hospitalization.

For each patient, the laboratory values recorded in the analytical dataset were included. The source dataset did not contain sufficiently detailed and consistently verified timestamps to establish the exact timing of blood collection relative to hospital admission, establishment of the CHF diagnosis, or administration of individual treatments. Therefore, the laboratory variables were interpreted as measurements documented during the index hospitalization and not specifically as pretreatment or admission measurements.

Inflammatory and Immune-Derived Biomarkers

The inflammatory and immune assessment included white blood cell count (WBC), absolute neutrophil

count (ANC), absolute lymphocyte count (ALC), absolute monocyte count (AMC), CRP, and fibrinogen.

Five cell count-derived inflammatory indices were calculated from the corresponding absolute blood cell counts. The neutrophil-to-lymphocyte ratio (NLR) was calculated as ANC/ALC, the platelet-to-lymphocyte ratio (PLR) as platelet count/ALC, and the monocyte-to-lymphocyte ratio (MLR) as AMC/ALC. The systemic immune-inflammation index (SII) was calculated as (ANC × platelet count)/ALC, and the systemic inflammation response index (SIRI) as (ANC × AMC)/ALC.

Because several of these indices share the same component cell counts, their interrelationships were considered when representative predictors were selected for the regression models.

Hematological Parameters

The hematological assessment comprised red blood cell count, hemoglobin concentration, hematocrit, mean corpuscular volume, red cell distribution width expressed as coefficient of variation (RDW-CV), platelet count, and mean platelet volume.

Anemia was defined according to sex-specific World Health Organization hemoglobin thresholds as hemoglobin <12 g/dL in women and <13 g/dL in men [25]. Elevated RDW-CV was defined as >14.5%.

Information regarding anemia subtype, ferritin, transferrin saturation, serum iron, vitamin B12, folate, active bleeding, recent transfusion, and volume status was not systematically available and could not be included in the analysis.

Renal, Metabolic, and Electrolyte Parameters

The renal, metabolic, and electrolyte assessment included serum creatinine, urea, estimated glomerular filtration rate (eGFR), blood glucose, sodium, potassium, and chloride.

The eGFR was calculated from the serum creatinine values recorded in the analytical dataset using the race-free 2021 Chronic Kidney Disease Epidemiology Collaboration creatinine equation, incorporating age and sex [26]. Reduced renal function was defined as eGFR <60 mL/min/1.73 m^2. Because eGFR was calculated from a single creatinine measurement documented during the index hospitalization, this threshold was not used to establish a diagnosis of chronic kidney disease.

Hyponatremia was defined as serum sodium <135 mmol/L, hypokalemia as serum potassium <3.5 mmol/L, and hyperkalemia as serum potassium >5.5 mmol/L.

Hepatic and Coagulation Parameters

The hepatic assessment included aspartate aminotransferase (AST), alanine aminotransferase (ALT), and the AST/ALT ratio.

The coagulation assessment included prothrombin time (PT), prothrombin activity, international normalized ratio (INR), activated partial thromboplastin time (APTT), the APTT ratio (patient APTT divided by the laboratory control APTT), and fibrinogen.

Although fibrinogen has both inflammatory and coagulation-related interpretations, it was assigned to the inflammatory family for multiplicity correction and was not treated as a second independent hypothesis when additionally presented with the coagulation parameters.

Comorbidities and pharmacological treatment

Clinical comorbidities were identified from diagnoses and other clinical information recorded in the electronic medical records. The evaluated comorbidities included arterial hypertension, type 2 diabetes mellitus, coronary artery disease, atrial fibrillation or flutter, previous myocardial infarction, previous percutaneous coronary intervention or coronary artery bypass grafting, documented chronic kidney disease, chronic pulmonary disease, chronic liver disease, previous stroke or transient ischemic attack, and malignancy.

Active infection, active autoimmune or inflammatory disease, and recent bleeding or transfusion were not used as formal exclusion criteria. These conditions were not evaluated using a standardized screening protocol applied to the entire cohort. Investigations such as urine cultures, screening swabs, or chest radiography were performed only when clinically indicated, and their results were not systematically extracted.

Information on medications documented during the index hospitalization was also extracted. The assessed

treatments included diuretics, mineralocorticoid receptor antagonists, sodium-glucose cotransporter 2 inhibitors, corticosteroids, antiplatelet therapy, vitamin K antagonists, direct oral anticoagulants, and unfractionated or low-molecular-weight heparin. Medication data reflected treatments recorded in the hospital records during the index hospitalization; chronic pre-admission therapy was not systematically documented in the analytical dataset and may have been underreported. This is suggested by the low documented use of oral anticoagulants, beta-blockers, and angiotensin-converting enzyme inhibitors despite the high prevalence of atrial fibrillation and heart failure in the cohort.

Because the temporal relationship between laboratory testing and administration of individual medications could not be established reliably, medication variables were analyzed descriptively and were not included as covariates in the primary regression models. Accordingly, the observed laboratory differences cannot be interpreted as necessarily preceding or being independent of treatment exposure.

Statistical analysis

Statistical analyses evaluated cross-sectional associations between laboratory values documented during the index hospitalization and CHF status recorded as a final discharge diagnosis during the same hospitalization. Descriptive statistics and between-group comparisons were performed using IBM SPSS Statistics, version 26.0 (IBM Corp., Armonk, NY). Conventional and Firth-penalized logistic regression, correlation, receiver operating characteristic (ROC), model-performance, and bootstrap analyses were performed using Python, version 3.12.14 (Python Software Foundation, Wilmington, DE), with pandas 2.2.3, NumPy 2.3.5, SciPy 1.17.0, and scikit-learn 1.8.0.

Continuous variables are presented as medians and interquartile ranges (Q1-Q3) and were compared between patients with and without CHF using the Mann-Whitney U test. Categorical variables are presented as numbers and percentages and were compared using Pearson's chi-square test or Fisher's exact test, as appropriate.

To account for multiple testing, the Benjamini-Hochberg procedure was applied separately to the inflammatory, hematological, renal/metabolic/electrolyte, and hepatic/coagulation families of variables. Fibrinogen was included only in the inflammatory multiplicity family. The same procedure was applied separately to the comorbidity and treatment variables. Multiplicity-adjusted results are reported as q-values, with $q < 0.05$ considered statistically significant.

Spearman rank correlation coefficients were calculated in all 107 patients to describe the relationships among biomarkers associated with CHF and to identify variables containing overlapping biological or mathematical information. The correlation analysis was interpreted descriptively and was not used as an automated variable-selection procedure.

Biomarkers showing significant between-group differences after multiplicity correction were subsequently evaluated in domain-specific logistic regression models. A restricted number of representative variables was selected to reduce redundancy among strongly correlated biomarkers and indices derived from the same laboratory measurements. All models were adjusted for age and female sex, with female sex coded as 1 and male sex used as the reference category.

The inflammatory model included CRP and NLR, whereas the hematological model included anemia and RDW-CV >14.5%. Because eGFR, creatinine, and urea were strongly correlated, they were evaluated in three separate renal models. The electrolyte model included hyponatremia, the hepatic model included ALT, and the coagulation model included INR and APTT. PT, prothrombin activity, and INR were not entered simultaneously because they are derived from the same coagulation assay and were strongly correlated, and the APTT ratio was not entered together with APTT.

Given the limited number of patients with hyponatremia, its association with CHF was additionally evaluated using a profile-likelihood confidence interval and Firth-penalized logistic regression. A profile-likelihood confidence interval was also calculated for eGFR <60 mL/min/1.73 m^2 because of its relatively large effect estimate. Because coagulation parameters could have been influenced by anticoagulant treatment, PT, INR, and APTT were additionally compared between patients with and without CHF in the subgroup of 73 patients without documented anticoagulant exposure.

In the domain-specific models, CRP and NLR were analyzed per one-unit increase, creatinine per 1 mg/dL, urea per 10 mg/dL, ALT per 10 U/L, INR per 0.1 unit, and APTT per one second. Linearity of continuous predictors on the logit scale was assessed using the Box-Tidwell procedure. Logistic regression results are reported as regression coefficients (β), standard errors, odds ratios, 95% confidence intervals, and two-sided p-values. Benjamini-Hochberg correction was additionally applied across the 11 biomarker terms evaluated in the domain-specific models; age and sex were not included in this correction.

Following the domain-specific analyses, a post hoc integrated model was constructed to represent the inflammatory, hematological, and renal domains central to the study objective. The model and its predictors

were not specified before data analysis; they were selected after review of the between-group comparisons and the domain-specific regression results, and the model is therefore exploratory. Age and female sex were retained as demographic adjustment variables. CRP was selected as a conventional and clinically interpretable marker of systemic inflammation, anemia as a clinically relevant hematological variable, and eGFR <60 mL/min/1.73 m^2 as a standardized indicator of reduced renal function. One representative variable from each domain was included to limit overlapping information and model complexity. No prespecified or automated selection algorithm, such as stepwise selection, was used. The resulting model included five predictor parameters: age, female sex, CRP, anemia, and eGFR <60 mL/min/1.73 m^2. In this model, CRP was expressed per 10 mg/L increase. With 49 patients with CHF, the model corresponded to 9.8 events per parameter; this ratio should not be interpreted as excluding overfitting or accounting for the preceding variable-selection process. Because of the limited number of CHF events, further adjustment for other clinically relevant factors, such as documented chronic kidney disease, diuretic or other treatment exposure, left ventricular ejection fraction, and hemodynamic status, was not undertaken. Such adjustment would have increased the risk of overfitting, and several of these variables were incompletely available or could not be temporally related to the laboratory measurements.

Six sensitivity analyses were performed to determine whether the principal findings depended on the selected biomarker or model specification. NLR replaced CRP in Sensitivity Model 1; RDW-CV >14.5% replaced anemia in Sensitivity Model 2; urea replaced eGFR <60 mL/min/1.73 m^2 in Sensitivity Model 3; creatinine replaced eGFR <60 mL/min/1.73 m^2 in Sensitivity Model 4; APTT was added to the post hoc integrated model in Sensitivity Model 5; and ALT was added in Sensitivity Model 6. Age and female sex were retained in all sensitivity models. These analyses were used to assess the robustness of the principal associations rather than to select an optimal predictive model.

Multicollinearity in the post hoc integrated model was assessed using variance inflation factors. Model fit and apparent performance were summarized using the Akaike information criterion (AIC), Nagelkerke R^2, area under the ROC curve, and Brier score. AIC values were compared only between models fitted to the same patients.

The 95% confidence intervals for the apparent AUCs were estimated using 3,000 stratified bootstrap resamples. AUC and AIC values for the sensitivity models were interpreted descriptively and were not used to select an optimal model.

The post hoc integrated model, with its predictor set fixed, underwent internal validation using 3,000 nonparametric bootstrap resamples. In each resample, the following steps were repeated: a sample of 107 patients was drawn with replacement from the original cohort; the logistic regression model was refitted with the same five predictors to re-estimate the coefficients; the AUC, Brier score, calibration intercept, and calibration slope were calculated in the bootstrap sample; and the refitted model was applied to the original cohort to calculate the same measures. The average difference between the bootstrap-sample and original-cohort estimates represented bootstrap optimism and was subtracted from the apparent estimates to obtain optimism-corrected values. Ideal values for the calibration intercept and slope are 0 and 1, respectively. The between-group comparisons, the domain-specific regression models, and the selection of the five predictors were not repeated within the bootstrap resamples. Consequently, this procedure does not capture the uncertainty introduced by predictor selection, and the optimism-corrected estimates may still overestimate the performance of the model.

All 107 patients had complete data for the variables included in the comparative and multivariable analyses; therefore, no missing-data imputation was performed. All statistical tests were two-sided. Statistical significance was defined as P <0.05 for unadjusted analyses and q <0.05 for multiplicity-adjusted analyses. Given the retrospective, cross-sectional design and post hoc model development, all regression and model-performance analyses were considered exploratory.

Ethical considerations

All data were anonymized before analysis. The study was approved by the Ethics Committee of Lucian Blaga University of Sibiu (Approval No. 2579/June 19, 2026) and was conducted in accordance with the principles of the Declaration of Helsinki.

Results

Baseline characteristics of the study population

Baseline demographic characteristics of patients with and without CHF are presented in Table *2*. No statistically significant differences between groups were observed regarding age, age category ($\chi^2(1) = 0.85$, $P = 0.355$, $\phi = 0.09$), sex distribution ($\chi^2(1) = 0.78$, $P = 0.378$, $\phi = 0.09$), or area of residence ($\chi^2(1) = 0.26$, $P = 0.609$, $\phi = 0.05$), indicating that the two groups were adequately balanced for subsequent comparisons.

Variable	Overall (*n* = 107)	Without CHF (*n* = 58)	With CHF (*n* = 49)	*P*-value
Age (years)	72.0 (66.0-78.0)	72.0 (68.0-77.8)	71.0 (65.0-78.0)	0.363
Age ≥ 65 years, *n* (%)	85 (79.44)	48 (82.76)	37 (75.51)	0.355 ($\chi^2(1) = 0.85$; $\varphi = 0.09$)
Sex, *n* (%)				0.378 ($\chi^2(1) = 0.78$; $\varphi = 0.09$)
Female	53 (49.53)	31 (53.45)	22 (44.90)	
Male	54 (50.47)	27 (46.55)	27 (55.10)	
Area of residence, *n* (%)				0.609 ($\chi^2(1) = 0.26$; $\varphi = 0.05$)
Rural	31 (28.97)	18 (31.03)	13 (26.53)	
Urban	76 (71.03)	40 (68.97)	36 (73.47)	

TABLE 2: Demographic characteristics of patients with severe MR according to CHF status.

Continuous data are presented as median (interquartile range (IQR)), and categorical data as number (*n*) and percentage (%). Between-group comparisons were performed using the Mann-Whitney U test for continuous variables and Pearson's chi-square test for categorical variables. $\chi^2(1)$, Pearson's chi-square statistic with one degree of freedom; φ, phi coefficient (effect size).

CHF, congestive heart failure; MR, mitral regurgitation

Comorbidities, documented medication use, echocardiographic findings, and anticoagulant exposure

As described in the Materials and Methods section, free-text echocardiographic data were retrievable for a subset of patients, with left ventricular ejection fraction numerically lower in patients with CHF (40% vs. 50%); quantitative regurgitant parameters and right ventricular function remained unavailable for systematic comparison between groups.

The distribution of comorbidities and medications documented during the index hospitalization is presented in Table *3*. After correction for multiple comparisons, documented chronic kidney disease was more frequent among patients with CHF than among those without CHF (36.7% vs. 10.3%; Fisher's exact test, $P = 0.002$, $\phi = 0.32$, $q = 0.023$). No other between-group difference in the assessed comorbidities remained statistically significant after correction.

Variable	Overall (*n* = 107)	Without CHF (*n* = 58)	With CHF (*n* = 49)	*P*-value	*q*-value
Comorbidities, *n* (%)					
Hypertension	74 (69.2%)	46 (79.3%)	28 (57.1%)	0.020 (φ = 0.24)	0.074
Type 2 diabetes mellitus	33 (30.8%)	15 (25.9%)	18 (36.7%)	0.294 (φ = 0.12)	0.359
Coronary artery disease	37 (34.6%)	16 (27.6%)	21 (42.9%)	0.108 (φ = 0.16)	0.150
Atrial fibrillation/flutter	67 (62.6%)	32 (55.2%)	35 (71.4%)	0.109 (φ = 0.17)	0.150
Prior myocardial infarction	16 (15.0%)	5 (8.6%)	11 (22.4%)	0.059 (φ = 0.19)	0.129
Prior PCI/CABG	22 (20.6%)	10 (17.2%)	12 (24.5%)	0.472 (φ = 0.09)	0.519
Chronic kidney disease (coded)	24 (22.4%)	6 (10.3%)	18 (36.7%)	0.002 (φ = 0.32)	0.023
COPD/chronic pulmonary disease	14 (13.1%)	4 (6.9%)	10 (20.4%)	0.047 (φ = 0.20)	0.129
Chronic liver disease	5 (4.7%)	0 (0.0%)	5 (10.2%)	0.018 (φ = 0.24)	0.074
Prior stroke/TIA	10 (9.3%)	5 (8.6%)	5 (10.2%)	1.000 (φ = 0.03)	1.000
Malignancy	6 (5.6%)	1 (1.7%)	5 (10.2%)	0.091 (φ = 0.18)	0.150
In-hospital treatment, *n* (%)					
Diuretics (loop)	41 (38.3%)	8 (13.8%)	33 (67.3%)	<0.001 (φ = 0.55)	<0.001
Mineralocorticoid receptor antagonists	25 (23.4%)	3 (5.2%)	22 (44.9%)	<0.001 (φ = 0.47)	<0.001
ACE inhibitors	8 (7.5%)	2 (3.4%)	6 (12.2%)	0.139 (φ = 0.17)	0.243
ARNI (sacubitril/valsartan)	7 (6.5%)	2 (3.4%)	5 (10.2%)	0.243 (φ = 0.14)	0.309
Beta-blockers	7 (6.5%)	1 (1.7%)	6 (12.2%)	0.046 (φ = 0.21)	0.107
SGLT2 inhibitors	17 (15.9%)	1 (1.7%)	16 (32.7%)	<0.001 (φ = 0.42)	<0.001
Vitamin K antagonists	2 (1.9%)	0 (0.0%)	2 (4.1%)	0.207 (φ = 0.15)	0.290
LMWH/unfractionated heparin	30 (28.0%)	9 (15.5%)	21 (42.9%)	0.002 (φ = 0.30)	0.008
Direct oral anticoagulants	2 (1.9%)	1 (1.7%)	1 (2.0%)	1.000 (φ = 0.01)	1.000
Antiplatelet agents	21 (19.6%)	9 (15.5%)	12 (24.5%)	0.329 (φ = 0.11)	0.356
Statins	6 (5.6%)	1 (1.7%)	5 (10.2%)	0.091 (φ = 0.18)	0.183
Iron therapy (IV)	2 (1.9%)	0 (0.0%)	2 (4.1%)	0.207 (φ = 0.15)	0.290
Antibiotics	4 (3.7%)	1 (1.7%)	3 (6.1%)	0.330 (φ = 0.12)	0.356
Corticosteroids	9 (8.4%)	1 (1.7%)	8 (16.3%)	0.011 (φ = 0.26)	0.030

TABLE 3: Comorbidities and medications documented during the index hospitalization according to congestive heart failure status.

Comparisons between patients with and without CHF were performed using Fisher's exact test; φ (phi coefficient) is reported as the effect size. *P*-values were adjusted for multiple comparisons separately within the comorbidity and medication domains using the Benjamini-Hochberg procedure, yielding *q*-values. Medication entries indicate treatment documented at any time during the index hospitalization. The available records did not permit reliable distinction among chronic pre-admission therapy, emergency treatment administered before laboratory sampling, and treatment initiated after sampling; these variables are therefore presented descriptively and are not interpreted as exposures preceding the admission laboratory measurements.

φ, phi coefficient; *n*, number of patients; CHF, congestive heart failure; PCI, percutaneous coronary intervention; CABG, coronary artery bypass grafting; COPD, chronic obstructive pulmonary disease; TIA, transient ischemic attack; ACE, angiotensin-converting enzyme; ARNI, angiotensin receptor-neprilysin inhibitor; SGLT2, sodium-glucose cotransporter 2; LMWH, low-molecular-weight heparin; IV, intravenous.

The medical records of patients with CHF more frequently documented the use of loop diuretics (67.3% vs. 13.8%; p<0.001, ϕ=0.55, q<0.001), mineralocorticoid receptor antagonists (44.9% vs. 5.2%; p<0.001, ϕ=0.47, q<0.001), sodium-glucose cotransporter 2 inhibitors (32.7% vs. 1.7%; p<0.001, ϕ=0.42, q<0.001), and corticosteroids (16.3% vs. 1.7%; p=0.011, ϕ=0.26, q=0.030), all assessed using Fisher's exact test.

Documented exposure to unfractionated or low-molecular-weight heparin was also more frequent among patients with CHF (42.9% vs. 15.5%; Fisher's exact test, p=0.002, ϕ=0.30, q=0.008), whereas no significant between-group differences were observed for vitamin K antagonists or direct oral anticoagulants. These data describe medications documented at any time during the index hospitalization and should not be interpreted as treatment effects or as exposures that necessarily preceded laboratory testing.

Inflammatory and immune-derived biomarker profile according to CHF status

Compared with patients without CHF, those with CHF exhibited a significantly altered inflammatory profile, characterized by higher CRP, ANC, NLR, MLR, SII, and SIRI together with lower ALC (Table 4). These differences remained statistically significant after Benjamini-Hochberg correction (all $q < 0.05$), whereas WBC, AMC, fibrinogen, and PLR did not differ significantly between the groups.

Biomarker	Overall (*n* = 107)	Without CHF (*n* = 58)	With CHF (*n* = 49)	*P*-value	*q*-value
CRP, mg/L	2.50 (1.15-8.85)	1.80 (1.00-3.95)	6.50 (1.30-45.30)	<0.001	0.002
NLR	2.37 (1.84-3.77)	2.17 (1.72-2.90)	3.15 (2.11-5.36)	<0.001	<0.001
ANC, ×10⁹/L	4.23 (3.45-6.06)	4.02 (3.24-5.46)	5.50 (3.73-7.32)	0.006	0.011
ALC, ×10⁹/L	1.73 (1.30-2.20)	1.84 (1.59-2.29)	1.51 (1.11-2.06)	0.006	0.011
MLR	0.35 (0.28-0.47)	0.31 (0.25-0.38)	0.40 (0.33-0.52)	<0.001	<0.001
SII, ×10⁹/L	545.65 (389.05-793.60)	479.39 (380.38-659.41)	623.27 (402.09-1097.86)	0.025	0.039
SIRI, ×10⁹/L	1.59 (1.02-2.59)	1.26 (0.92-1.89)	2.31 (1.26-3.25)	<0.001	<0.001
WBC, ×10⁹/L	7.12 (5.97-8.96)	6.79 (5.85-8.50)	7.90 (6.23-10.11)	0.063	0.086
AMC, ×10⁹/L	0.63 (0.50-0.76)	0.61 (0.49-0.73)	0.66 (0.53-0.85)	0.142	0.156
PLR	114.88 (91.75-149.11)	110.93 (92.66-149.14)	115.24 (91.67-148.21)	0.689	0.691
Fibrinogen, mg/dL	352.60 (298.70-407.45)	337.55 (275.62-401.27)	368.00 (305.50-424.60)	0.099	0.121

TABLE 4: Inflammatory and immune-derived biomarkers according to CHF status.

Data are presented as median (interquartile range (IQR)). Comparisons were performed using the Mann-Whitney U test. P-values were adjusted for multiple comparisons within the inflammatory domain using the Benjamini-Hochberg procedure, yielding *q*-values.

CRP, C-reactive protein; NLR, neutrophil-to-lymphocyte ratio; ANC, absolute neutrophil count; ALC, absolute lymphocyte count; MLR, monocyte-to-lymphocyte ratio; SII, systemic immune-inflammation index; SIRI, systemic inflammation response index; WBC, white blood cell count; AMC, absolute monocyte count; PLR, platelet-to-lymphocyte ratio; CHF, congestive heart failure; *n*, number of patients

Hematological profile according to CHF status

Compared with patients without CHF, those with CHF showed a significantly higher prevalence of anemia and higher red cell distribution width (RDW-CV) values (Table 5). Although median hemoglobin levels did not differ significantly between groups, the proportion of patients meeting criteria for anemia was more than five-fold higher in the CHF group. Similarly, both RDW-CV as a continuous variable and the proportion of patients with elevated RDW-CV (>14.5%) were significantly higher among patients with CHF. Platelet count was lower in patients with CHF before correction for multiple comparisons but was no longer statistically significant after Benjamini-Hochberg correction. RBC count, hemoglobin, hematocrit, MCV, and MPV did not differ significantly between the groups.

Hematological parameter	Overall (*n* = 107)	Without CHF (*n* = 58)	With CHF (*n* = 49)	*P*-value	*q*-value
Anemia, *n* (%)	22 (20.56)	4 (6.89)	18 (36.73)	<0.001 ($\chi^2(1) = 14.48$; φ = 0.37)	<0.001
RDW-CV >14.5%, *n* (%)	35 (32.71)	10 (17.24)	25 (51.02)	<0.001 ($\chi^2(1) = 13.77$; φ = 0.36)	<0.001
RDW-CV, %	13.40 (12.60-15.15)	13.10 (12.50-14.35)	14.60 (13.10-16.20)	0.001	0.004
RBC count, $\times 10^{12}$/L	4.54 (4.19-5.05)	4.57 (4.39-4.84)	4.54 (3.75-5.21)	0.689	0.778
Hb, g/dL	14.00 (12.70-15.35)	14.05 (13.10-14.70)	13.70 (11.60-15.90)	0.630	0.778
Hct, %	40.50 (37.20-43.95)	40.50 (38.15-42.58)	39.70 (35.40-46.30)	0.691	0.778
MCV, fL	89.20 (85.05-91.65)	89.30 (85.98-90.50)	88.80 (83.70-92.90)	0.854	0.854
PLT, $\times 10^9$/L	209.00 (163.50-255.00)	222.50 (178.50-261.50)	199.00 (129.00-247.00)	0.027	0.060
MPV, fL	10.90 (10.40-11.60)	10.90 (10.40-11.50)	11.10 (10.40-11.70)	0.483	0.778

TABLE 5: Hematological parameters according to CHF status.

Continuous variables are presented as median (interquartile range (IQR)) and categorical variables as number (*n*) and percentage (%). Comparisons were performed using the Mann-Whitney U test for continuous variables and Pearson's chi-square test for categorical variables. The variables in this table constituted a single family of hypotheses for Benjamini-Hochberg correction, and adjusted results are reported as *q*-values. Anemia was defined as hemoglobin <12 g/dL in women or <13 g/dL in men. Elevated RDW-CV was defined as >14.5%. $\chi^2(1)$, Pearson's chi-square statistic with one degree of freedom.

φ, phi coefficient (effect size); RDW-CV, red cell distribution width coefficient of variation; RBC, red blood cell; Hb, hemoglobin; Hct, hematocrit; MCV, mean corpuscular volume; PLT, platelet count; MPV, mean platelet volume; CHF, congestive heart failure

Renal, metabolic, and electrolyte profile according to CHF status

Patients with CHF showed significantly worse renal function compared with those without CHF (Table *6*). Urea and creatinine were markedly higher, and eGFR was markedly lower, in the CHF group. The proportion of patients with eGFR <60 mL/min/1.73 m^2 was higher in the CHF group than in the group without CHF (61.22% vs. 20.69%). This threshold was used to indicate reduced renal function documented during the index hospitalization and not to establish a diagnosis of chronic kidney disease. Hyponatremia was also significantly more frequent in the CHF group. Glucose and hypokalemia differed between the groups before correction for multiple comparisons but were no longer statistically significant after Benjamini-Hochberg correction. Potassium, sodium, and chloride concentrations and the prevalence of hyperkalemia did not differ significantly between the groups.

Parameter	Overall (*n* = 107)	Without CHF (*n* = 58)	With CHF (*n* = 49)	*P*-value	*q*-value
eGFR, mL/min/1.73 m²	71.02 (47.61-91.05)	80.31 (64.24-93.43)	50.20 (31.00-79.42)	<0.001	<0.001
eGFR <60 mL/min/1.73 m², *n* (%)	42 (39.25)	12 (20.69)	30 (61.22)	<0.001 ($\chi^2(1) = 18.30$; $\varphi = 0.41$)	<0.001
Creatinine, mg/dL	1.00 (0.84-1.30)	0.89 (0.76-1.05)	1.25 (0.99-2.09)	<0.001	<0.001
Urea, mg/dL	45.00 (34.01-69.08)	38.01 (30.00-48.05)	60.00 (45.00-96.00)	<0.001	<0.001
Blood glucose, mg/dL	110.50 (97.02-138.50)	114.50 (101.08-142.37)	101.06 (94.84-130.30)	0.038	0.070
Sodium, mmol/L	141.00 (139.00-142.07)	141.00 (139.04-142.08)	140.02 (137.04-142.00)	0.172	0.210
Hyponatremia, *n* (%)	11 (10.28)	2 (3.44)	9 (18.37)	0.011 ($\chi^2(1) = 6.41$; $\varphi = 0.24$)	0.025
Potassium, mmol/L	4.40 (4.00-4.60)	4.40 (4.10-4.60)	4.40 (3.90-4.60)	0.410	0.451
Hypokalemia, *n* (%)	5 (4.67)	1 (1.72)	4 (8.16)	0.170 (Fisher; $\varphi = 0.15$)	0.210
Hyperkalemia, *n* (%)	4 (3.74)	3 (5.17)	1 (2.04)	0.626 (Fisher; $\varphi = 0.08$)	0.626
Chloride, mmol/L	107.07 (104.03-109.06)	107.01 (105.31-108.82)	107.02 (102.00-109.70)	0.111	0.174

TABLE 6: Renal, metabolic, and electrolyte profile according to CHF status.

Continuous variables are presented as median (interquartile range (IQR)) and categorical variables as number (*n*) and percentage (%). Comparisons were performed using the Mann-Whitney U test for continuous variables, Pearson's chi-square test for eGFR <60 mL/min/1.73 m² and hyponatremia, and Fisher's exact test for hypokalemia and hyperkalemia. The variables in this table constituted a single family of hypotheses for Benjamini-Hochberg correction, and adjusted results are reported as *q*-values. eGFR was calculated using the 2021 Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) creatinine equation. Hyponatremia was defined as sodium <135 mmol/L, hypokalemia as potassium <3.5 mmol/L, and hyperkalemia as potassium >5.5 mmol/L. $\chi^2(1)$, Pearson's chi-square statistic with one degree of freedom.

φ, phi coefficient (effect size); Fisher, Fisher's exact test; eGFR, estimated glomerular filtration rate; CHF, congestive heart failure

Hepatic and coagulation profile according to CHF status

In the comparison of patients with severe MR with and without CHF, coagulation abnormalities were more pronounced than hepatic alterations. Patients with CHF had higher PT, INR, and APTT values and lower prothrombin activity than patients without CHF (Table 7). These differences remained statistically significant after Benjamini-Hochberg correction. Because PT, prothrombin activity, and INR are derived from the same coagulation assay and are mathematically related, and because anticoagulant use could confound these findings, we performed a sensitivity analysis restricted to the 73 patients without documented anticoagulant exposure (25 with CHF, 48 without). In this subgroup, PT, INR, and APTT remained significantly higher in patients with CHF (median PT 12.9 vs. 12.0 s; median INR 1.09 vs. 1.02; median APTT 31.3 vs. 28.9 s; all $P < 0.01$). However, because chronic oral anticoagulant therapy may have been underreported, this subgroup may have included patients receiving anticoagulant treatment, and the analysis cannot exclude an effect of anticoagulation. Among the hepatic biomarkers, ALT was higher in patients with CHF, whereas AST, the AST/ALT ratio, and fibrinogen did not differ significantly between the groups after correction.

Parameter	Overall (*n* = 107)	Without CHF (*n* = 58)	With CHF (*n* = 49)	*P*-value	*q*-value
PT, s	12.50 (11.90-13.80)	12.10 (11.50-12.67)	13.40 (12.50-15.20)	<0.001	<0.001
Prothrombin activity, %	90.50 (74.30-98.80)	96.10 (87.03-106.53)	76.30 (62.20-92.60)	<0.001	<0.001
INR	1.06 (1.01-1.19)	1.02 (0.97-1.08)	1.16 (1.05-1.30)	<0.001	<0.001
APTT ratio	1.02 (0.93-1.24)	0.97 (0.87-1.03)	1.18 (1.02-1.45)	<0.001	<0.001
APTT, s	30.50 (27.80-37.20)	29.10 (26.20-30.80)	35.70 (30.50-45.10)	<0.001	<0.001
AST, U/L	23.00 (17.75-27.25)	23.00 (17.00-25.75)	26.00 (19.25-32.00)	0.084	0.108
ALT, U/L	18.00 (14.75-27.25)	17.50 (13.00-21.75)	23.00 (17.00-31.00)	0.003	0.004
AST/ALT ratio	1.23 (0.96-1.62)	1.30 (1.06-1.60)	1.08 (0.93-1.65)	0.182	0.182

TABLE 7: Hepatic and coagulation parameters according to CHF status.

Data are presented as median (interquartile range (IQR)). Comparisons were performed using the Mann-Whitney U test. The variables in this table constituted a single family of hypotheses for Benjamini-Hochberg correction, and adjusted results are reported as *q*-values. The APTT ratio is the ratio of the patient's APTT to the laboratory control APTT. Laboratory reference ranges were: PT, 9.9-12.3 s; prothrombin activity, 76.6%-116.2%; INR, 0.86-1.1; APTT ratio, 0-1.2; APTT, 25.1-37.7 s.

PT, prothrombin time; INR, international normalized ratio; APTT ratio, activated partial thromboplastin time ratio; APTT, activated partial thromboplastin time; AST, aspartate aminotransferase; ALT, alanine aminotransferase; s, seconds; CHF, congestive heart failure; *n*, number of patients

Correlation analysis between inflammatory, hematological, renal, and coagulation biomarkers

Spearman correlation analysis (*n* = 107 for all pairwise correlations) revealed three key patterns of association among biomarkers significantly related to CHF (Figure *2*). First, the prothrombin-based indices were very strongly intercorrelated (PT vs. INR, rho = 0.98; INR vs. prothrombin activity, rho = -0.99; PT vs. prothrombin activity, rho = -0.97), consistent with their derivation from the same clotting assay. The APTT ratio correlated almost perfectly with APTT (rho = 0.99), as expected from its derivation, whereas APTT correlated strongly with the prothrombin-based indices (rho = 0.75 with PT, 0.78 with INR, and -0.79 with prothrombin activity). Second, renal function markers showed the expected strong inverse relationship (creatinine vs. eGFR, rho = -0.92; urea vs. eGFR, rho = -0.74), while urea and creatinine correlated strongly with each other (rho = 0.75). Third, within the inflammatory domain, NLR, SIRI, SII, and ANC were strongly intercorrelated (rho = 0.71-0.86), reflecting their shared dependence on the absolute neutrophil count, whereas ALC showed the expected inverse relationship with these neutrophil-driven indices. CRP and ALT showed comparatively weak correlations with other biomarkers, suggesting they capture at least partially distinct pathophysiological information. RDW-CV showed moderate, statistically significant correlations across multiple domains, including with NLR (rho = 0.47) and SII (rho = 0.34) within the inflammatory cluster, with urea (rho = 0.44) and eGFR (rho = -0.35) within the renal cluster, and with INR and APTT within the coagulation cluster, suggesting that RDW-CV reflects a biological signal shared across several systems rather than a single isolated pathway.

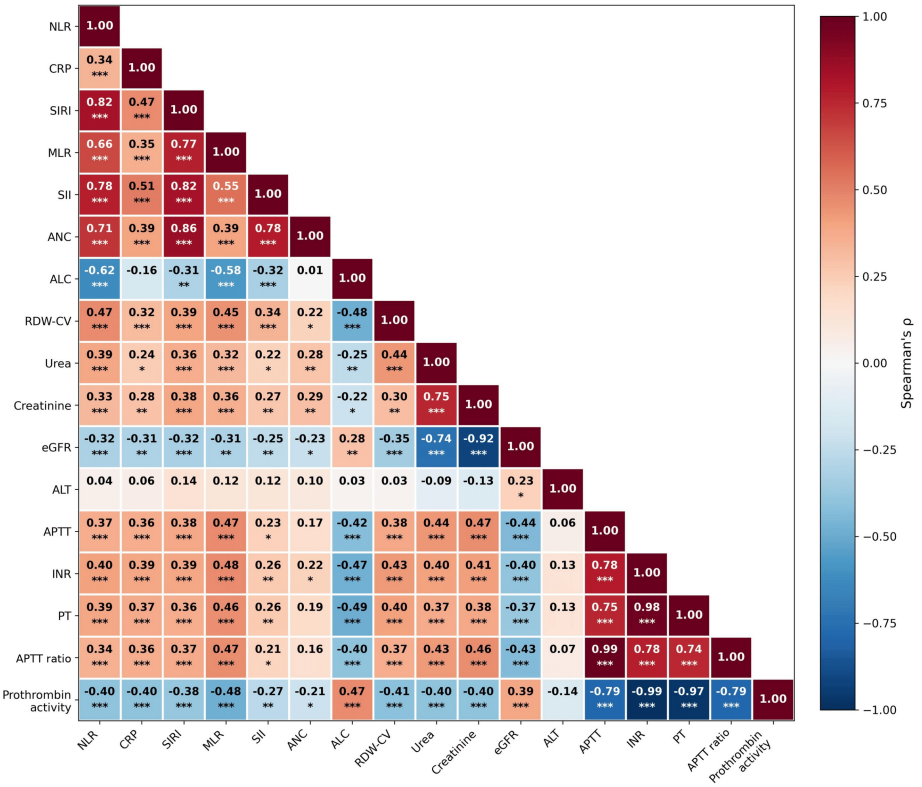


FIGURE 2: Correlation matrix of biomarkers associated with CHF in severe MR.

Values represent Spearman's rank correlation coefficients (ρ) calculated in all 107 patients. *P < 0.05; **P < 0.01; ***P < 0.001; cells without asterisks indicate $P \geq 0.05$.

The image was generated using Python, version 3.12.14 (Python Software Foundation, Wilmington, DE), with the open-source libraries Matplotlib (version 3.11) and Seaborn (version 0.13.2).

ρ, Spearman's rank correlation coefficient; CHF, congestive heart failure; MR, mitral regurgitation; NLR, neutrophil-to-lymphocyte ratio; CRP, C-reactive protein; SIRI, systemic inflammation response index; MLR, monocyte-to-lymphocyte ratio; SII, systemic immune-inflammation index; ANC, absolute neutrophil count; ALC, absolute lymphocyte count; RDW-CV, red cell distribution width coefficient of variation; eGFR, estimated glomerular filtration rate; ALT, alanine aminotransferase; APTT, activated partial thromboplastin time; INR, international normalized ratio; PT, prothrombin time; APTT ratio, activated partial thromboplastin time ratio

Domain-specific multivariable models

Across the six biological domains, multivariable logistic regression models adjusted for age and female sex identified associations between several biomarkers and CHF (Table *8*). In the inflammatory model, both NLR (OR 1.32, 95% CI 1.01-1.71; $P = 0.039$) and CRP (OR 1.03, 95% CI 1.00-1.07; $P = 0.026$) were associated with CHF after adjustment for age and female sex. In the hematological model, both anemia (OR 3.61, 95% CI 1.21-10.77; $P = 0.022$) and RDW-CV >14.5% (OR 3.88, 95% CI 1.50-10.02; $P = 0.005$) were associated with CHF after adjustment for age and female sex.

Model	Term	β	SE	OR (95% CI)	*P*-value	*q*-value
1. Inflammatory	Intercept	-0.245	1.558	-	0.875	
	Age, per year	-0.013	0.022	0.99 (0.95-1.03)	0.535	
	Female sex	-0.575	0.451	0.56 (0.23-1.36)	0.202	
	NLR, per unit	0.275	0.133	1.32 (1.01-1.71)	0.039	0.048
	CRP, per unit	0.034	0.015	1.03 (1.00-1.07)	0.026	0.036
2. Hematological	Intercept	0.010	1.472	-	0.994	

	Age, per year	-0.010	0.021	0.99 (0.95-1.03)	0.639	
	Female sex	-0.433	0.439	0.65 (0.27-1.53)	0.324	
	Anemia	1.283	0.558	3.61 (1.21-10.77)	0.022	0.035
	RDW-CV >14.5%	1.355	0.484	3.88 (1.50-10.02)	0.005	0.018
3. Renal 1	Intercept	1.505	1.507	-	0.318	
	Age, per year	-0.030	0.022	0.97 (0.93-1.01)	0.163	
	Female sex	-0.871	0.474	0.42 (0.17-1.06)	0.066	
	eGFR < 60 mL/min/1.73 m² [a]	2.237	0.504	9.37 (3.48-25.17)	<0.001	<0.001
3. Renal 2	Intercept	-0.154	1.415	-	0.913	
	Age, per year	-0.009	0.020	0.99 (0.95-1.03)	0.653	
	Female sex	-0.317	0.406	0.73 (0.33-1.61)	0.435	
	Creatinine, per mg/dL	0.587	0.254	1.80 (1.09-2.96)	0.021	0.035
3. Renal 3	Intercept	-0.546	1.541	-	0.723	
	Age, per year	-0.028	0.023	0.97 (0.93-1.02)	0.207	
	Female sex	-0.469	0.467	0.63 (0.25-1.56)	0.315	
	Urea, per 10 mg/dL	0.494	0.128	1.64 (1.28-2.11)	<0.001	<0.001
4. Electrolyte	Intercept	0.373	1.386	-	0.788	
	Age, per year	-0.007	0.019	0.99 (0.96-1.03)	0.719	
	Female sex	-0.458	0.408	0.63 (0.28-1.41)	0.262	
	Hyponatremia [b]	1.941	0.818	6.97 (1.40-34.64)	0.018	0.035
4. Electrolyte (Firth-corrected) [c]	Hyponatremia	1.741	0.777	5.70 (1.24-26.15)	0.025	
5. Hepatic	Intercept	-0.571	1.496	-	0.703	
	Age, per year	-0.001	0.020	1.00 (0.96-1.04)	0.961	
	Female sex	-0.466	0.405	0.63 (0.28-1.39)	0.250	
	ALT, per 10 U/L	0.310	0.155	1.36 (1.01-1.85)	0.046	0.051
6. Coagulation	Intercept	-4.711	2.158	-	0.029	
	Age, per year	-0.021	0.022	0.98 (0.94-1.02)	0.344	
	Female sex	-0.449	0.469	0.64 (0.25-1.60)	0.338	
	INR, per 0.1 unit	0.216	0.172	1.24 (0.89-1.74)	0.209	0.209
	APTT, per second	0.114	0.046	1.12 (1.02-1.23)	0.013	0.035

TABLE 8: Multivariable logistic regression models for biomarkers associated with CHF in patients with severe MR.

All models were adjusted for age and sex (*n* = 107). ORs are reported with Wald 95% CIs; *q*-values reflect Benjamini-Hochberg correction for multiple testing applied across the 11 biomarker terms evaluated in this table. Linearity of continuous predictors on the logit scale was confirmed using the Box-Tidwell test (all *P* > 0.05). Multicollinearity within each domain-specific model was assessed using variance inflation factors, with all values below 2, indicating no evidence of problematic collinearity.

[a]Profile-likelihood (likelihood-ratio) 95% CI: 3.67-26.65; conclusions were unchanged relative to the Wald CI reported above.

[b]Profile-likelihood (likelihood-ratio) 95% CI: 1.66-47.32; conclusions were unchanged relative to the Wald CI reported above.

[c]Firth-penalized logistic regression, performed as a bias-correction sensitivity analysis given the limited number of hyponatremia events (*n* = 11).

β, regression coefficient; CHF, congestive heart failure; MR, mitral regurgitation; *n*, number of patients; CI, confidence interval; OR, odds ratio; SE, standard error; NLR, neutrophil-to-lymphocyte ratio; CRP, C-reactive protein; RDW-CV, red cell distribution width coefficient of variation; eGFR, estimated glomerular filtration rate; INR, international normalized ratio; APTT, activated partial thromboplastin time; ALT, alanine aminotransferase

Among the three renal models, eGFR < 60 mL/min/1.73 m² showed the strongest association (OR 9.37, 95% CI 3.48-25.17; *P* < 0.001), followed by creatinine (OR 1.80, 95% CI 1.09-2.96; *P* = 0.021) and urea (OR 1.64 per 10 mg/dL, 95% CI 1.28-2.11; *P* < 0.001). Because the Wald confidence interval for eGFR <60 was comparatively wide, a profile-likelihood confidence interval was additionally computed (3.67-26.65), confirming the conclusion of the Wald-based estimate.

In the electrolyte model, hyponatremia was associated with CHF (OR 6.97, 95% CI 1.40-34.64; *P* = 0.018); given the limited number of hyponatremia events (*n* = 11), a profile-likelihood confidence interval (1.66-47.32) and a Firth-penalized sensitivity analysis were performed, yielding a more conservative but still statistically significant estimate (OR 5.70, 95% CI 1.24-26.15; *P* = 0.025). In the hepatic model, ALT was associated with CHF (OR 1.36 per 10 U/L, 95% CI 1.01-1.85; *P* = 0.046). In the coagulation model, APTT was significantly associated with CHF (OR 1.12 per second, 95% CI 1.02-1.23; *P* = 0.013), whereas INR was not (*P* = 0.209).

Because these 11 biomarker terms were evaluated across separate domain-specific models, Benjamini-Hochberg correction was applied across all terms to account for multiple testing. After correction, ALT no longer reached statistical significance (*q* = 0.051), whereas all other previously significant associations remained significant (*q* ⩽ 0.048), including NLR, CRP, anemia, RDW-CV, eGFR < 60 mL/min/1.73 m², creatinine, urea, hyponatremia, and APTT. Age and female sex were not significantly associated with CHF in any of the eight domain-specific models. Linearity of continuous predictors on the logit scale was confirmed using the Box-Tidwell test (all P > 0.05), supporting the validity of the linear logistic specification used throughout.

Post hoc integrated model and sensitivity analyses

In the post hoc integrated model, eGFR < 60 mL/min/1.73 m² was associated with CHF (OR 5.56, 95% CI 1.94-15.94; *P* = 0.001) after adjustment for age, female sex, CRP, and anemia (Table *9*). CRP showed a positive but statistically imprecise association (OR 1.34, 95% CI 0.99-1.83; *P* = 0.062), and neither age, female sex, nor anemia reached statistical significance after mutual adjustment. The post hoc integrated model demonstrated acceptable apparent discrimination (AUC 0.809, 95% CI 0.725-0.884; AIC 124.0).

Analysis	Variable/model specification	β	SE	OR (95% CI)	*P*-value	AIC	Apparent AUC (95% CI)
Post hoc integrated model	Intercept	0.965	1.587	2.63	0.543	124.0	0.809 (0.725-0.884)
	Age, per year	-0.026	0.023	0.97 (0.93-1.02)	0.244		
	Female sex	-0.833	0.498	0.43 (0.16-1.15)	0.094		
	CRP, per 10 mg/L	0.296	0.159	1.34 (0.99-1.83)	0.062		
	Anemia	0.561	0.656	1.75 (0.48-6.35)	0.392		
	eGFR < 60 mL/min/1.73 m²	1.715	0.538	5.56 (1.94-15.94)	0.001		
Sensitivity 1	NLR replacing CRP: NLR, per unit	0.294	0.134	1.34 (1.03-1.74)	0.028	121.4	0.812 (0.722-0.887)
	eGFR < 60 mL/min/1.73 m²	1.830	0.546	6.23 (2.14-18.17)	0.001		
Sensitivity 2	RDW-CV > 14.5% replacing anemia	1.016	0.526	2.76 (0.98-7.75)	0.054	121.0	0.821 (0.734-0.894)
	eGFR <60 mL/min/1.73 m²	1.683	0.546	5.38 (1.85-15.68)	0.002		
Sensitivity 3	Urea replacing eGFR: urea, per 10 mg/dL	0.445	0.132	1.56 (1.20-2.02)	0.001	113.5	0.857 (0.778-0.923)
Sensitivity 4	Creatinine, per mg/dL	0.210	0.258	1.23 (0.74-2.05)	0.416	134.3	0.781 (0.693-0.861)
Sensitivity 5	APTT added to post hoc integrated model: APTT, per second	0.100	0.041	1.10 (1.02-1.20)	0.014	117.2	0.837 (0.756-0.909)
	eGFR < 60 mL/min/1.73 m²	1.219	0.581	3.38 (1.08-10.57)	0.036		
Sensitivity 6	ALT added to post hoc integrated model: ALT, per 10 U/L	0.289	0.221	1.33 (0.87-2.06)	0.191	123.2	0.809 (0.722-0.887)
	eGFR < 60 mL/min/1.73 m²	1.885	0.583	6.59 (2.10-20.63)	0.001		

TABLE 9: Post hoc integrated logistic regression model and sensitivity analyses for CHF in patients with severe MR.

All models were fitted in 107 patients. AUC values represent apparent discrimination in the development cohort, and their 95% CIs were estimated using 3,000 stratified bootstrap resamples. AIC and apparent AUC values for the sensitivity models are presented descriptively and should not be interpreted as demonstrating superiority of one model over another. Bootstrap internal validation and optimism-corrected performance estimates for the post hoc integrated model are reported separately in the following table.

β, regression coefficient; SE, standard error; OR, odds ratio; CI, confidence interval; AIC, Akaike information criterion; AUC, area under the receiver operating characteristic curve; CRP, C-reactive protein; eGFR, estimated glomerular filtration rate; NLR, neutrophil-to-lymphocyte ratio; RDW-CV, red cell distribution width coefficient of variation; APTT, activated partial thromboplastin time; ALT, alanine aminotransferase; CHF, congestive heart failure; MR, mitral regurgitation

Reduced eGFR remained associated with CHF in all four sensitivity models in which it was retained, with odds ratios ranging from 3.38 (95% CI 1.08-10.57; Sensitivity Model 5) to 6.59 (95% CI 2.10-20.63; Sensitivity Model 6). This association was maintained when alternative inflammatory or hematological variables were used and when APTT or ALT was added to the post hoc integrated model. Substituting urea for eGFR < 60 mL/min/1.73 m² (Sensitivity Model 3) yielded the numerically lowest AIC (113.5) and highest apparent AUC (0.857, 95% CI 0.778-0.923) among all models evaluated. These estimates were interpreted descriptively and

do not establish superior performance relative to the post hoc integrated model or other sensitivity models.

In contrast, when creatinine replaced eGFR < 60 (Sensitivity Model 4), creatinine was not significantly associated with CHF (OR 1.23, 95% CI 0.74-2.05; $P = 0.416$), and the model had an AIC of 134.3 and an apparent AUC of 0.781. Thus, the renal findings differed according to the marker used to represent renal function. When APTT was added to the post hoc integrated model (Sensitivity Model 5), both APTT (OR 1.10, 95% CI 1.02-1.20; $P = 0.014$) and eGFR < 60 mL/min/1.73 m² remained statistically significant in the model. When ALT was added (Sensitivity Model 6), eGFR < 60 remained significant, whereas ALT did not (OR 1.33, 95% CI 0.87-2.06; $P = 0.191$).

Collectively, reduced eGFR remained the most consistent renal marker associated with CHF in the post hoc integrated model and in all sensitivity models in which it was retained. Higher urea was also associated with CHF when used as the alternative renal marker, whereas creatinine was not. These findings indicate that the renal association was supported by eGFR and urea but was not consistent across all renal markers.

Performance and internal validation of the fixed post hoc integrated model

The post hoc integrated model had an apparent AUC of 0.809 (95% CI 0.725-0.884). After bootstrap correction for optimism, the AUC was 0.780 (Table *10*). The apparent Brier score was 0.175, and the optimism-corrected Brier score was 0.196. The optimism-corrected calibration intercept was -0.008, while the calibration slope of 0.797 indicated some degree of overfitting. The Nagelkerke R^2 was 0.378, and variance inflation factors ranged from 1.07 to 1.47, providing no evidence of problematic multicollinearity. These estimates apply only to the fixed post hoc integrated model specification and do not account for uncertainty introduced by the preceding biomarker-screening and model-development process.

Performance measure	Apparent estimate	Optimism-corrected estimate
AUC	0.809	0.780
Brier score	0.175	0.196
Calibration intercept	0.000	-0.008
Calibration slope	1.000	0.797
Nagelkerke R^2	0.378	-
VIF range	1.07-1.47	-

TABLE 10: Apparent and bootstrap-corrected performance of the fixed post hoc integrated logistic regression model.

The validated model corresponds to the post hoc integrated model presented in Table 9 and included age, female sex, CRP, anemia, and eGFR < 60 mL/min/1.73 m². The model was fitted in 107 patients, including 49 patients with CHF. The 95% CI for the apparent AUC was estimated using 3,000 stratified bootstrap resamples. Optimism correction for the AUC, Brier score, calibration intercept, and calibration slope was performed through a separate internal-validation procedure using 3,000 bootstrap resamples. Nagelkerke R^2 and VIF values represent apparent estimates and were not corrected for optimism. The bootstrap procedure evaluated the fixed predictor specification and did not repeat the preceding biomarker-screening or variable-selection process.

AUC, area under the receiver operating characteristic curve; CI, confidence interval; VIF, variance inflation factor; CRP, C-reactive protein; eGFR, estimated glomerular filtration rate; CHF, congestive heart failure

Discussion

This retrospective study compared routinely available laboratory measures across six biological domains in 107 hospitalized patients with severe MR, stratified by a documented concomitant diagnosis of heart failure. The main finding was not the identification of a new biomarker, but the differing consistency of laboratory associations across domains. In age- and sex-adjusted domain-specific models, inflammatory, hematological, renal, electrolyte, and coagulation variables were associated with heart failure status after multiplicity correction. However, when representative inflammatory, hematological, and renal measures were examined together in a post hoc model, reduced eGFR was the only included biomarker retaining statistical significance. This finding should be understood as a cross-sectional association rather than evidence that renal dysfunction independently causes heart failure or adds predictive value beyond established clinical and echocardiographic assessment. Interpretation is further limited by potential confounding from pre-existing kidney disease, heart failure therapy, and incomplete characterization of MR etiology and ventricular function. Nevertheless, because eGFR is routinely measured and renal function

directly influences heart failure therapy and procedural risk assessment, this association is clinically relevant to the evaluation of patients with severe MR, as discussed below.

Previous investigations have described biomarker patterns in patients with heart failure and examined their relationships with concomitant MR [4,23]. These studies compared patients with heart failure with and without MR, whereas the present study was restricted to patients with echocardiographically documented severe MR and compared them according to concomitant CHF status. To our knowledge, the combined assessment of inflammatory, hematological, and renal laboratory parameters in this specific population has not been extensively reported. The contribution of the present study therefore lies in three aspects: the restriction to a cohort with severe MR, allowing laboratory differences to be examined according to CHF status rather than according to the presence of MR; the concurrent evaluation of multiple routinely available laboratory domains with domain-specific multiplicity correction; and the examination of whether domain-specific associations persisted when representative inflammatory, hematological, and renal measures were analyzed together. This approach showed that reduced eGFR was the most consistent correlate of CHF, whereas inflammatory and hematological associations were attenuated after mutual adjustment. Our findings should therefore be interpreted as an extension of the descriptive evidence to a cohort restricted to severe MR, rather than as the identification of previously unknown heart failure biomarkers. Because natriuretic peptide results were not systematically available, we could not assess whether routine laboratory measures provided information beyond BNP or NT-proBNP.

Because all patients in this cohort had severe MR, and no comparison group of patients with heart failure without MR was available, the present study cannot determine which of the observed laboratory abnormalities are specific to severe MR. The overall pattern of reduced renal function, inflammatory activation, anemia, and hyponatremia is similar to that described in general heart failure populations, and none of the observed associations should therefore be regarded as MR-specific [4,23]. What the study design does allow is a description of the laboratory abnormalities that accompany concomitant CHF among patients who share the same severe valvular lesion, thereby reducing heterogeneity related to the presence and severity of MR. Severe MR may plausibly contribute to these abnormalities through chronic volume overload, elevated pulmonary venous pressures, and systemic congestion, but this pathway could not be distinguished from other causes of heart failure in our data. Moreover, because MR etiology and mechanism were not consistently available, we could not examine whether the laboratory profile differed between primary and secondary MR. Comparative studies including patients with heart failure without MR and patients with non-severe MR are needed to identify any MR-specific laboratory features.

Patients with CHF in our cohort had higher CRP, ANC, NLR, MLR, SII, and SIRI and lower ALC. This pattern is compatible with increased innate immune activation and relative lymphocyte depletion, as previously described in chronic heart failure. However, these inflammatory abnormalities are nonspecific and may also reflect comorbidity, physiological stress, or the overall severity of illness [6,7,9,18,20,27]. Treatment exposure should also be considered. Corticosteroid use was documented more frequently among patients with CHF (16.3% vs. 1.7%), and corticosteroids increase circulating neutrophil counts and decrease lymphocyte counts. Part of the observed differences in NLR, SII, SIRI, and related indices may therefore reflect steroid exposure rather than heart failure-related inflammation, particularly because the timing of administration relative to blood sampling could not be established.

In age- and sex-adjusted models, both CRP and NLR were associated with documented CHF. The CRP association was attenuated in the post hoc integrated model, whereas NLR remained associated with CHF when substituted for CRP in a sensitivity analysis. Associations between systemic inflammatory indices and heart failure have also been reported in population-based studies [16,28]. In our cohort, the differences between individual markers may reflect overlapping biological information, residual confounding, and limited precision rather than distinct causal roles. The absence of a difference in PLR may be explained by the parallel reduction in platelet and lymphocyte counts among patients with CHF, which leaves the ratio largely unchanged; the same platelet effect may account for the smaller difference observed for SII compared with the other neutrophil-based indices.

Beyond inflammation, anemia was markedly more prevalent and RDW-CV significantly higher among patients with CHF, consistent with the multifactorial origin of anemia in heart failure, including chronic inflammation, impaired iron metabolism, reduced erythropoietin production, and hemodilution [29,30]. RDW-CV correlated significantly with inflammatory (NLR, SII), renal (urea, eGFR), and coagulation (INR, APTT) markers, and elevated RDW has previously been linked to increased mortality in hospitalized patients with CHF [31]. In the hematological model, both anemia and RDW-CV > 14.5% were associated with CHF. In the post hoc integrated model, anemia retained a positive but imprecise association, with a confidence interval including unity, and RDW-CV > 14.5% was similarly imprecise when it replaced anemia in the sensitivity analysis. Thus, anemia and erythrocyte anisocytosis characterize the hematological profile of CHF, but their distinct contributions cannot be separated reliably in this cohort. Because anemia etiology could not be classified, iron, vitamin B12, or folate deficiency, bleeding, impaired erythropoietin production, inflammation, and congestion-related hemodilution may all have contributed. Anemia should therefore be interpreted as a concurrent hematological finding rather than as an antecedent risk factor for CHF.

Reduced eGFR emerged as the strongest and most consistent correlate of CHF and remained associated with

CHF after adjustment for age, female sex, CRP, and anemia in the post hoc integrated model. This finding is compatible with cardiorenal interactions involving reduced renal perfusion, venous congestion, renin-angiotensin-aldosterone system activation, and systemic inflammation [5]. Because eGFR was calculated from a single creatinine measurement obtained during hospitalization, values below 60 mL/min/1.73 m^2 may reflect acute kidney injury or worsening renal function secondary to congestion, hypoperfusion, or treatment, rather than stable chronic kidney disease. Baseline pre-admission creatinine values were not available in the analytical dataset to distinguish these conditions. Urea was also associated with CHF, both in the renal model and when it replaced eGFR in the post hoc integrated model, and this specification showed the most favorable apparent fit among all models. A plausible explanation is that urea is particularly sensitive to neurohormonal activation and renal congestion. In states of reduced effective circulating volume, renin-angiotensin-aldosterone system activation and non-osmotic vasopressin release enhance tubular reabsorption of sodium and water, and urea reabsorption increases in parallel, so that urea may rise disproportionately to creatinine. Urea may therefore integrate information on renal function, hemodynamic status, and neurohormonal activation in heart failure. However, urea is also influenced by loop diuretic therapy, catabolism, and protein intake, and the differences in model fit were interpreted descriptively and do not establish the superiority of this model. In contrast, creatinine was not associated with CHF when it replaced eGFR in the integrated model. eGFR was selected for the post hoc integrated model as a standardized and clinically interpretable measure of reduced renal function, whereas creatinine is additionally influenced by muscle mass and nutritional status. The persistence of a renal association when renal status was represented by either reduced eGFR or increased urea, but not by creatinine, supports the relevance of cardiorenal dysfunction in this population, while indicating that the choice of renal marker materially affects model performance. Nevertheless, this finding should be interpreted as a statistical association rather than evidence of a causal relationship. Renal function may also reflect disease severity, hemodynamic status, comorbidities, and administered treatments. Documented chronic kidney disease and diuretic exposure were more frequent among patients with CHF, and neither these factors nor ventricular function or hemodynamic status could be included in the adjusted models. Moreover, because the timing of blood sampling relative to admission, CHF diagnosis, and treatment could not be established, these cross-sectional findings cannot determine whether renal dysfunction preceded CHF or resulted from its hemodynamic and therapeutic consequences.

From a clinical perspective, reduced renal function was common in this population, particularly among patients with concomitant CHF. Because eGFR is routinely available, it may help identify patients with severe MR in whom systemic involvement accompanying CHF is more likely and in whom closer monitoring of renal function, volume status, and electrolytes is warranted. Renal function is also directly relevant to the management of these patients. It influences the choice and dosing of heart failure therapies such as diuretics, mineralocorticoid receptor antagonists, and SGLT2 inhibitors, and it is a recognized component of operative risk assessment before surgical or transcatheter mitral intervention [32]. However, these considerations are based on cross-sectional associations. Our data do not show that eGFR improves risk stratification beyond established clinical and echocardiographic assessment, and its clinical usefulness in this setting requires prospective evaluation.

Hyponatremia was more frequent among patients with CHF and was associated with CHF in the domain-specific model, including in the Firth-penalized analysis. This finding is compatible with non-osmotic vasopressin release, neurohormonal activation, impaired water excretion, and diuretic exposure in heart failure [33]. However, hyponatremia was present in only 11 patients, and the wide confidence interval indicates substantial uncertainty regarding the magnitude of the association.

These renal and electrolyte findings should also be interpreted in light of treatment exposure. Loop diuretics (67.3% vs. 13.8%), mineralocorticoid receptor antagonists (44.9% vs. 5.2%), and SGLT2 inhibitors (32.7% vs. 1.7%) were documented considerably more frequently among patients with CHF. Loop diuretics may increase urea and creatinine through intravascular volume depletion and reduced renal perfusion and may contribute to hyponatremia. Mineralocorticoid receptor antagonists may increase serum creatinine and affect sodium and potassium concentrations. SGLT2 inhibitors are associated with an initial, generally reversible decline in eGFR. Because these medications were documented at any time during hospitalization and the timing of their administration relative to blood sampling could not be established, they were not included as covariates. The observed associations of eGFR, urea, creatinine, and hyponatremia with CHF may therefore partly reflect treatment effects rather than heart failure alone. The lower frequency of coded hypertension among patients with CHF may reflect coding practices or lower blood pressure in heart failure rather than a true difference in hypertensive disease.

Hepatic and coagulation abnormalities further characterized the multisystem involvement in CHF. Among hepatic markers, ALT was higher in patients with CHF and was nominally associated with CHF in the hepatic model, although this association did not remain significant after Benjamini-Hochberg correction, and it was no longer associated with CHF when added to the post hoc integrated model, in which reduced eGFR remained significant. The ALT elevation may be compatible with hepatic congestion or impaired hepatic perfusion, although ALT is nonspecific and may also reflect pre-existing hepatic disease, medication exposure, or other comorbid conditions, and its association with CHF in this cohort should be regarded as exploratory rather than robust.

Patients with CHF also exhibited prolonged PT, INR, APTT, and APTT ratio and reduced prothrombin activity. In the coagulation model, APTT remained associated with CHF, whereas INR did not. After APTT was added to the post hoc integrated model, APTT and reduced eGFR both remained associated with CHF. This finding requires particular caution because unfractionated or low-molecular-weight heparin exposure was more frequent among patients with CHF, and the timing of anticoagulant administration relative to blood sampling could not be established consistently. Notably, PT, INR, and APTT remained higher in the CHF group within the subgroup of 73 patients without documented anticoagulant exposure. This persistence is consistent with a possible contribution of hepatic congestion, reduced hepatic synthetic function, or endothelial dysfunction related to disease severity. However, documented oral anticoagulant use was low despite the high prevalence of atrial fibrillation, suggesting that chronic anticoagulant therapy may have been underreported; this subgroup may therefore have included anticoagulated patients. Consequently, the observed coagulation differences may reflect a combination of treatment exposure, hepatic congestion, and disease severity, and an intrinsic coagulopathy related to CHF cannot be established from these data [34].

Taken together, these findings indicate that CHF in severe MR is associated with inflammatory, hematological, renal, electrolyte, hepatic, and coagulation abnormalities. The discrimination of the post hoc integrated model decreased after bootstrap correction for optimism. Because the predictors were selected after review of the exploratory results and the bootstrap procedure did not repeat this selection, the optimism-corrected estimates do not incorporate the uncertainty arising from predictor selection, and the model's performance may still be overestimated. The internally validated calibration slope of 0.797 suggested some degree of overfitting, while the comparatively wide confidence interval around the apparent AUC indicated limited precision of the discrimination estimate. AUC and AIC values varied across the sensitivity models but were interpreted descriptively. These analyses were intended to examine the principal associations under alternative model specifications and do not establish the superiority of any model. The integrated model is exploratory and should not be regarded as a validated clinical tool or as ready for clinical use.

Because this is a cross-sectional study, these findings demonstrate that laboratory abnormalities are associated with concomitant CHF; they do not establish their temporal sequence or causal role. Reverse causation and residual confounding by unmeasured comorbidities, treatments, and cardiac characteristics remain plausible explanations. The biomarkers should therefore be interpreted as concurrent features of the clinical phenotype rather than predictors of future CHF development. Prospective external validation is required before the post hoc integrated model can be considered for clinical risk assessment. Because LVEF overestimates left ventricular function in severe MR, global longitudinal strain may detect left ventricular dysfunction before LVEF declines and has been associated with exercise capacity in asymptomatic patients with primary MR and preserved LVEF [35]. Future studies could therefore pair the inflammatory markers examined here with biventricular strain assessment.

Limitations

This study has several limitations. First, the retrospective, single-center, cross-sectional design and modest sample size (107 patients; 49 with CHF) limit statistical precision, generalizability, and causal interpretation. Second, CHF status relied on discharge diagnoses without independent adjudication; acute and chronic CHF were not consistently distinguished, and NYHA functional class and natriuretic peptides were not systematically available. Third, because only patients with severe MR were included, the study cannot determine whether the observed associations are specific to severe MR. In addition, inclusion required MR to be the principal diagnosis of the admission, which excluded patients admitted primarily for heart failure with severe secondary MR and limits the findings to patients admitted for their valve disease. The reason for admission (elective vs. acute) and admission vital signs were not extracted into the analytical dataset, and differences in admission type may account for some of the laboratory differences.

Fourth, echocardiographic reports were retrievable for 51 of 107 patients, and MR mechanism, detailed ventricular function, and quantitative parameters were incompletely available. Because LVEF overestimates left ventricular function in severe MR, and an LVEF ⩽ 60% is regarded as left ventricular dysfunction in primary MR [32], some patients without a CHF diagnosis may have had left ventricular dysfunction, which would reduce the contrast between the groups. Fifth, chronic pre-admission medication use, including oral anticoagulants, may have been underreported, and the timing of blood sampling relative to admission, CHF diagnosis, and treatment could not be established. Therefore, it cannot be determined whether laboratory abnormalities preceded CHF or reflected clinical status and treatment during hospitalization. Sixth, adjustment was limited to age and sex and, additionally, to CRP and anemia in the integrated model. The limited number of CHF events did not permit adjustment for documented chronic kidney disease, treatment exposure, ventricular function, or hemodynamic status. Seventh, eGFR was derived from a single in-hospital creatinine measurement and may reflect acute kidney injury rather than chronic kidney disease. Finally, the integrated model was developed post hoc in the same cohort. Bootstrap validation of the fixed predictor set did not repeat predictor selection and may underestimate optimism. No external validation was performed; therefore, predictive utility and incremental clinical value remain unestablished.

Conclusions

Among 107 hospitalized patients with severe MR, documented concomitant CHF was associated with abnormalities across inflammatory, hematological, renal, electrolyte, hepatic, and coagulation domains. Reduced eGFR showed the most consistent association with CHF in exploratory multivariable analyses, whereas several other associations weakened after mutual adjustment. As a routinely available measure, eGFR may help identify patients with severe MR in whom systemic involvement accompanying CHF is more likely. Given the cross-sectional, single-center design and incomplete cardiac phenotyping, these findings are hypothesis-generating and require confirmation in prospective studies incorporating NYHA functional class, NT-proBNP, and standardized echocardiographic phenotyping, including MR mechanism and quantification (effective regurgitant orifice area and regurgitant volume), left ventricular and left atrial volumes, right ventricular function, and myocardial strain.

Additional Information

Author Contributions

All authors have reviewed the final version to be published and agreed to be accountable for all aspects of the work.

Concept and design: Nastaca Alina Palade, Emilia Sorina Fiat, Crina Cristina Solomon, Razvan Constantin Vonica, Bogdan Ioan Vintila, Felicia Gabriela Gligor

Acquisition, analysis, or interpretation of data: Nastaca Alina Palade, Emilia Sorina Fiat, Crina Cristina Solomon, Razvan Constantin Vonica, Bogdan Ioan Vintila, Maria Totan, Anca Maria Arseniu , Claudiu Morgovan, Anca Butuca , Adina Frum, Carmen Maximiliana Dobrea, Felicia Gabriela Gligor, Diter Atasie

Drafting of the manuscript: Nastaca Alina Palade, Emilia Sorina Fiat, Crina Cristina Solomon

Critical review of the manuscript for important intellectual content: Nastaca Alina Palade, Emilia Sorina Fiat, Crina Cristina Solomon, Razvan Constantin Vonica, Bogdan Ioan Vintila, Maria Totan, Anca Maria Arseniu , Claudiu Morgovan, Anca Butuca , Adina Frum, Carmen Maximiliana Dobrea, Felicia Gabriela Gligor, Diter Atasie

Supervision: Felicia Gabriela Gligor

Disclosures

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Data are available on reasonable request. The data are stored as de-identified patient data, which are available on request to Nastaca Alina Palade (nastacaalina.coman@ulbsibiu.ro).

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