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ORIGINAL RESEARCH

Association of Hematological Biomarkers of Inflammation with 10-Year Major Adverse Cardiovascular Events and All-Cause Mortality in Patients with Metabolic Dysfunction-Associated Steatotic Liver Disease: The ARIC Study

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Background: Metabolic dysfunction-associated steatotic liver disease (MASLD) increases the risk of cardiovascular disease and existing evidence indicates that MASLD affects the cardiovascular system through systemic inflammation. Our aim was to assess the association of hematological biomarkers of inflammation with the 10-year risk of major adverse cardiovascular events (MACE) and all-cause mortality in MASLD patients.

Methods: A total of 1858 MASLD participants from the Atherosclerosis Risk in Communities cohort study at visit 2 (1990–1992) were included. A total of 1338 non-MASLD participants were also included in the comparison. At baseline, hematological biomarkers of inflammation such as leukocytes, neutrophils, lymphocytes, monocytes, and C-reactive protein (CRP) were measured. Participants were followed up for MACE and all-cause mortality for a period of 10 years. Multivariate adjusted Cox models were used to estimate hazard ratios (HR).

Results: The 10-year MACE was higher in MASLD participants than in non-MASLD participants (20.8% vs 9.3%). Monocytes (HR 1.114, [95% CI, 1.022–1.216] per 1-SD, P=0.015) and CRP (HR 1.109 [95% CI, 1.032–1.190] per 1-SD, P=0.005) were associated with an increased 10-year risk of MACE, independent of other cardiovascular risk factors. This association was specific to the MASLD population. None of these hematological biomarkers demonstrated a significant association with 10-year all-cause mortality.

Conclusion: Increased levels of monocytes and CRP were associated with an increased 10-year risk of MACE in the MASLD population. Hematological biomarkers of inflammation may help identify MASLD populations at higher risk for cardiovascular events.

Keywords: MASLD, biomarkers, inflammation, cardiovascular

Introduction

Non-alcoholic fatty liver disease (NAFLD) is currently the most prevalent chronic liver disease worldwide, with a global prevalence of approximately 25% of the adult population.[1] NAFLD is a multisystem disease where insulin resistance and related metabolic dysfunction play a pathogenic role in the development of NAFLD and its liver-related complications and extrahepatic complications, such as cardiovascular disease, type 2 diabetes mellitus, and chronic kidney disease.[2,3] Consequently, three large pan-national liver associations proposed that metabolic dysfunction-associated steatotic liver disease (MASLD) should replace the term NAFLD.[4] MASLD is associated with an increased prevalence and incidence of cardiovascular disease, which is the main cause of mortality in

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MASLD patients.[2,5–8] Besides insulin resistance, inflammation is also believed to play a significant role in driving the progression of MASLD, and existing evidence indicates that MASLD may affect the cardiovascular system via systemic inflammation.[8–10] Systemic inflammation can be measured using various biochemical or hematological markers commonly obtained from routine blood tests.[11] Specifically, the roles of leukocytes, their subtypes, and C-reactive protein (CRP) have been utilized to quantify systemic inflammation.[12,13]

Currently, the relationship between systemic inflammation and future cardiovascular adverse events in patients with MASLD remains unclear. Understanding this association can help us better understand the value of hematological parameters as risk markers and the potential of anti-inflammatory therapy in reducing future cardiovascular events in the MASLD population. Several anti-inflammatory drugs for treating non-alcoholic steatohepatitis (NASH) are currently undergoing clinical trials.[7,9,10] Thus, our study aimed to assess the association of various hematological biomarkers of inflammation with the 10-year risk of major adverse cardiovascular events (MACE) and all-cause mortality in MASLD patients, by leveraging data from the Atherosclerosis Risk in Communities (ARIC) study.

Methods

Study Population

The ARIC study is a community-based cohort that includes 15,792 individuals aged 45 to 64 recruited from four communities (Washington County, Maryland; Forsyth County, North Carolina; Minneapolis, MN; and Jackson, MS) in the United States during visit 1 (1987–1989).[14] To investigate the association between hematological biomarkers of inflammation and MASLD, we included participants who attended the ARIC visit 2 (1990–1992). We excluded participants with missing data on gamma-glutamyl transferase (GGT), body mass index (BMI), triglycerides, and waist circumference. Subsequently, we used a validated algorithm to define MASLD (see MASLD, below), then we categorized the remaining participants into MASLD group and non-MASLD group. Among the MASLD group, we further excluded those who reported heavy alcohol consumption (see MASLD, below). For the analysis of the outcomes, we excluded participants with missing hematological data at visit 2 from both groups. After these exclusions, the non-MASLD group consisted of 1338 ARIC participants, and the MASLD group consisted of 1858 ARIC participants (Figure 1).

In this study, visit 2 of the ARIC was considered as the baseline. All participants underwent a maximum of 10 years of follow-up from the baseline examination or until the occurrence of disease/death or loss to follow-up.

MASLD

MASLD was defined as the presence of hepatic steatosis (detected either by biopsy, imaging Methods, or blood biomarkers/scores) and no other liver etiologies (including consuming heavy alcohol) coupled with at least one of the following criteria: a fasting plasma glucose (FPG) ≥100 mg/dL or a history of hypoglycemic medication use, overweight or obesity (waist girth>94cm for male and waist girth>80cm for female or BMI ≥ 25 kg/m^2), blood pressure (BP) ≥ 130/85 mmHg or specific drug treatment, high-density lipoprotein cholesterol (HDL-C) ≤40 mg/dL for male and ≤50 mg/dL for female or lipid lowering treatment, plasma triglycerides ≥ 150 mg/dL or lipid lowering treatment.[4] Since visit 2 of the ARIC did not include liver imaging or histological results, the presence of hepatic steatosis was diagnosed based on a fatty liver index (FLI) ≥60 (Table S1),[5,6,15] which has been externally validated in the general population or in grade 3 obese persons.[5,15] For large epidemiologic studies, whenever imaging tools are not available or feasible, FLI was described as an acceptable alternative for the diagnosis of steatosis by the European Clinical Practice Guidelines.[5] Heavy alcohol consumption was defined as consuming more than one drink per day for female and more than two drinks per day for male.[6] Non-MASLD group was diagnosed based on a FLI<30. We used the Fibrosis-4 index (FIB-4) (Table S1) to assess the degree of liver fibrosis, with a threshold of FIB-4≥2.67 reflecting a high probability of advanced fibrosis.[5,6,16,17]

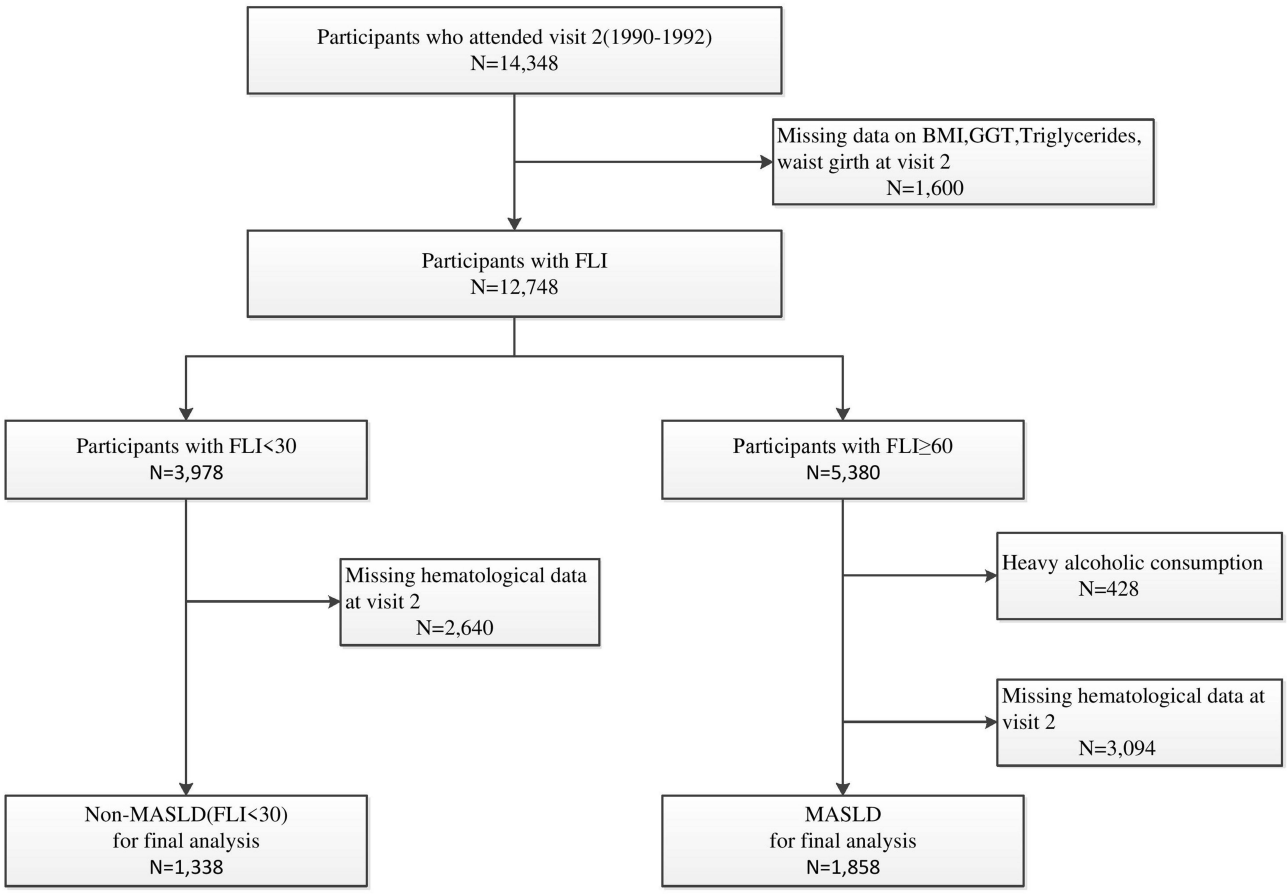

Figure 1 Flowchart of participants selection.
Abbreviations: ARIC, Atherosclerosis Risk in Communities; BMI, Body mass index; GGT, Gamma-glutamyl transferase; MASLD, Metabolic Dysfunction-associated Steatotic Liver Disease.

Covariates

Sex, race, age, smoking, alcohol use, and medications use were self-reported. Height, weight, and waist girth were measured by trained personnel. BMI was calculated by dividing weight in kilograms by height in meters squared (kg/m^2). A certified technician measured the participants' BP three times, and the second and third measurements were averaged. Diabetes was defined as self-reported, diagnosed by a physician, or the use of hypoglycemic medications. Laboratory values were obtained from blood drawn at study visits.[18] Alanine transaminase (ALT), aspartate transaminase (AST), GGT, and CRP levels were measured in ARIC visit 2 (1990 to 1992) serum samples (stored at −80°C since collection) using the Roche Modular P Chemistry analyzer and reagents from Roche Diagnostics. Leukocytes were retrieved from whole anticoagulated blood, and total leukocyte count and differential leukocyte count (percentages) were determined by automated particle Coulter Counters within 24 hours after venipuncture in local hospital hematology laboratories. Absolute differential counts were computed from total leukocyte count and differential percentages. The modified hexokinase/glucose-6-phosphate dehydrogenase method was used to measure the blood glucose concentrations. Estimated glomerular filtration rate (eGFR) was calculated using the Chronic Kidney Disease Epidemiology Collaboration equation.[19]

Outcome Definition

The primary outcome of this study was the composite of all-cause mortality and MACE, which is defined as a combination of coronary heart disease (CHD), stroke, heart failure, or cardiovascular mortality.[20–22] Information about these outcomes was ascertained through active surveillance of local hospital discharge records, state death records,

linkage to the National Death Index, and annual phone interviews with participants or proxies. CHD was defined as definite or probable hospitalized myocardial infarction or fatal coronary heart disease.[22,23] Incident stroke was defined as definite or probable ischemic or hemorrhagic stroke.[20,24] Incident heart failure was defined as hospitalization or death based on the International Classification of Disease-9/10 code for heart failure (428, I50).[21] Cardiovascular mortality was defined as death with an International Classification of Disease-9/10 code of 390–459 or I00–I99 as the underlying cause of death.[23] The secondary outcome included CHD, stroke, heart failure, and cardiovascular mortality.

Statistical Analysis

Categorical variables were presented as counts and percentages (%). Continuous variables with normal distribution were described using means and standard deviations, whereas continuous variables with abnormal distribution were described using medians and interquartile ranges (IQR). Pearson Chi-squared test was used for categorical variables. Mann–Whitney *U*-test or Student's *t*-test was used to compare continuous variables as appropriate. Initially, the studied hematological biomarkers were considered as continuous variables, and we assessed the impact of a 1-SD increase in each biomarker on the 10-year risk of MACE and all-cause mortality. We used multivariate adjusted Cox regression models to estimate the hazard ratios (HR) of the outcome. Restricted cubic splines (RCS) were also used to flexibly model and visualization of hematological biomarkers in relation to 10-year MACE and all-cause mortality. We considered the following two models: model 1 was adjusted for age, sex, and race, and model 2 was further adjusted for BMI, diabetes, current smoking, systolic BP, diastolic BP, eGFR, total glucose, total cholesterol, triglycerides, HDL-C, and history of cardiovascular events. For sensitivity analysis, we restricted the analysis to MASLD participants not using lipid-lowering medications, which included 1709 MASLD participants not taking such medications. We restricted the analysis to MASLD participants with FIB-4<2.67 to assess the impact of the degree of fibrosis on the results alone which included 1814 participants with an FIB-4<2.67. Finally, we restricted the analysis to participants with MASLD free of a history of cardiovascular events. For a comparison, we further investigated the association between hematological biomarkers of inflammation and outcomes in the non-MASLD group. Statistical analyses were performed using the SPSS version 26.0 and R version 4.1.0, and statistical significance was inferred as a P value of <0.05.

Results

During ARIC visit 2, a total of 1858 MASLD participants and 1338 non-MASLD participants were included in the study. Compared to non-MASLD participants, those with MASLD had a higher proportion of males than females, and were more likely to have diabetes, obesity, and higher levels of total cholesterol, triglycerides, total glucose, systolic BP, diastolic BP, CRP, neutrophils, monocytes, lymphocytes, and leukocytes, while HDL-C was lower. The 10-year MACE was higher in MASLD participants than in the non-MASLD participants (20.8% vs 9.3%) (Table 1).

RCS analysis suggested that the association of hematological biomarkers with 10-year MACE and all-cause mortality typically followed a linear relationship (P for nonlinear>0.05) (Figures S1 and S2). After adjustment for demographic and cardiovascular risk factors, neutrophils, monocytes, and CRP level were positively associated with 10-year MACE (Table 2). Neutrophils, monocytes, CRP, and leukocytes were also significantly associated with 10-year all-cause mortality (P<0.05) (Table 2). We conducted sensitivity analyses by restricting the analysis to participants with MASLD who did not use lipid-lowering medications. Similar Results were observed in the sensitivity analysis (Table 3). Restricting the analysis to MASLD participants with FIB-4<2.67 rendered similar results, except that there was no longer an association between neutrophils and 10-year MACE, and monocytes were no longer associated with 10-year all-cause mortality (Table 4). When restricting the analysis to MASLD participants without a history of cardiovascular events, only monocytes and CRP remained associated with 10-year MACE, and no statistical association was found between all these hematological biomarkers and 10-year all-cause mortality (P >0.05) (Table 5). Furthermore, the associations were generally consistent across the subgroups (Figures S3 and S4). For comparison, we further investigated the association of monocytes and CRP with 10-year MACE in the non-MASLD group. In the non-MASLD group, RCS analysis suggested that the association of monocytes and CRP with 10-year MACE also followed a linear relationship (P for nonlinear>0.05) (Figure S5). No statistically significant association was found between monocytes (HR 0.979 [95% CI, 0.825–1.161] per 1-SD, P=0.807) and CRP (HR 0.881 [95% CI, 0.725–1.071] per 1-SD, P=0.202) and 10-year

Table 1 Baseline Characteristics of ARIC Study Participants at Visit 2

Characteristics	MASLD group (n=1858)	Non-MASLD group (n=1338)	*P* value
Age, Medians (IQR), years	57 (9)	56.5 (10)	0.034
Sex			<0.001
Female (%)	822 (44.2)	1026 (76.7)	
Male (%)	1036 (55.8)	312 (23.3)	
Race			0.018
Black (%)	139 (7.5)	72 (5.4)	
White (%)	1719 (92.5)	1266 (94.6)	
Body mass index, kg/m2	31.1 (5.5)	23.3 (2.4)	<0.001
Current smoking (%)	386 (20.8)	405 (30.3)	<0.001
Diabetes (%)	445 (24.0)	40 (3.0)	<0.001
Total Cholesterol, Medians (IQR), mg/dL	211 (51)	201 (44)	<0.001
Triglycerides, Medians (IQR), mg/dL	159.5 (104)	92 (51)	<0.001
HDL-C, Medians (IQR), mg/dl	10 (6)	16 (12)	<0.001
Total glucose, Medians (IQR), mg/dL	108 (22)	98 (11)	<0.001
eGFR, Medians (IQR), mL/min/1.73m2	95.5 (16.2)	97.9 (14.5)	<0.001
Systolic BP, Medians (IQR), mmHg	124 (21)	112 (21)	<0.001
Diastolic BP, Medians (IQR), mmHg	73 (13)	68 (13)	<0.001
History Cardiovascular Event (%)	327 (17.6)	119 (8.9)	<0.001
Leukocytes, Medians (IQR), 10^9/L	6.3 (2.1)	5.7 (2.2)	<0.001
Neutrophils, Medians (IQR), 10^9/L	3.8 (1.6)	3.5 (1.8)	<0.001
Lymphocytes, Medians (IQR), 10^9/L	1.9 (0.7)	1.7 (0.6)	<0.001
Monocytes, Medians (IQR), 10^9/L	0.4 (0.2)	0.3 (0.2)	<0.001
C-Reactive Protein, Medians (IQR), mg/L	3.5 (4.8)	2.3 (2.8)	<0.001
Major adverse cardiovascular events (%)	386 (20.8)	124 (9.3)	<0.001
Coronary outcomes (%)	275 (14.8)	75 (5.6)	<0.001
Stroke (%)	61 (3.3)	21 (1.6)	0.003
Heart failure (%)	132 (7.1)	39 (2.9)	<0.001
Cardiovascular mortality (%)	59 (3.2)	27 (2.0)	0.046
All-cause mortality (%)	173 (9.3)	115 (8.6)	0.485

Abbreviations: ARIC, Atherosclerosis Risk in Communities; BP, blood pressure; eGFR, estimated glomerular filtration rate; HDL-C, high-density lipoprotein cholesterol; MASLD, Metabolic Dysfunction-associated Steatotic Liver Disease.

MACE in the non-MASLD group (Table S2). The association of monocytes (HR 1.114 [95% CI, 1.022–1.216] per 1-SD, *P*=0.015) and CRP (HR 1.109 [95% CI, 1.032–1.190] per 1-SD, *P*=0.005) with 10-year MACE was specific to the MASLD group (Table 2). In the analysis of secondary outcomes, compared with non-MASLD group, monocytes and CRP levels were mainly associated with CHD and cardiovascular mortality in the MASLD group (Tables S3 and S4).

Discussion

In this cohort study involving 1858 MASLD participants, we found that increased levels of monocytes and CRP were associated with increased 10-year MACE risk, independent of other cardiovascular risk factors. This association was specific to the MASLD population. These findings were largely consistent across the subgroups. In the secondary outcome analysis, monocytes and CRP levels primarily showed an association with CHD and cardiovascular mortality. The results obtained from the sensitivity analyses argued a strong influence of the history of cardiovascular events on the study results.

To the best of our knowledge, no longitudinal study has evaluated the hematological biomarkers of inflammation for adverse cardiovascular outcomes in individuals with MASLD. To date, convincing epidemiological evidence indicates that MASLD is an independent cardiovascular risk factor.[3,5,25] And inflammation is an important driver in the pathogenesis of MASLD.[9,10] Previous studies have attempted to explain the relationship between inflammation and

Table 2 Hazard Ratios of 10-Year Major Adverse Cardiovascular Events and All-Cause Mortality from MASLD Group

	Model 1 HR (95% CI)	*P* value	Model 2 HR (95% CI)	*P* value
Major adverse cardiovascular events				
Leukocytes	1.211 (1.104–1.328)	<0.001	1.094 (0.985–1.215)	0.093
Neutrophils	1.236 (1.126–1.356)	<0.001	1.125 (1.015–1.249)	0.025
Lymphocytes	1.023 (0.928–1.128)	0.649	0.934 (0.845–1.033)	0.186
Monocytes	1.113 (1.026–1.207)	0.01	1.114 (1.022–1.216)	0.015
C-reactive protein	1.112 (1.046–1.183)	0.001	1.109 (1.032–1.190)	0.005
All-cause mortality				
Leukocytes	1.403 (1.236–1.593)	<0.001	1.226 (1.062–1.415)	0.005
Neutrophils	1.432 (1.264–1.623)	<0.001	1.267 (1.098–1.462)	0.001
Lymphocytes	1.077 (0.932–1.245)	0.313	0.974 (0.842–1.126)	0.718
Monocytes	1.163 (1.045–1.294)	0.005	1.153 (1.017–1.308)	0.026
C-reactive protein	1.148 (1.069–1.232)	<0.001	1.136 (1.048–1.231)	0.002

Notes: Model 1: Adjusted for age, sex, race, Model 2: Adjusted for age, sex, race, BMI, diastolic blood pressure, systolic blood pressure, eGFR, total glucose, total cholesterol, triglycerides, HDL-C, diabetes, current smoking, and history of cardiovascular events.
Abbreviations: ARIC, Atherosclerosis Risk in Communities; BMI, Body mass index; eGFR, estimated glomerular filtration rate; HDL-C, high-density lipoprotein cholesterol; MASLD, Metabolic Dysfunction-associated Steatotic Liver Disease.

Table 3 Hazard Ratios of 10-Year Major Adverse Cardiovascular Events and All-Cause Mortality from MASLD Group Not Using Lipid-Lowering Medications

	Model 2 HR (95% CI)	*P* value
Major adverse cardiovascular events		
Leukocytes	1.105 (0.986–1.238)	0.087
Neutrophils	1.133 (1.012–1.267)	0.030
Lymphocytes	0.939 (0.845–1.044)	0.244
Monocytes	1.132 (1.040–1.233)	0.004
C-reactive protein	1.097 (1.016–1.185)	0.019
All-cause mortality		
Leukocytes	1.270 (1.086–1.484)	0.003
Neutrophils	1.278 (1.097–1.490)	0.002
Lymphocytes	1.024 (0.882–1.188)	0.759
Monocytes	1.176 (1.040–1.330)	0.010
C-reactive protein	1.125 (1.034–1.225)	0.006

Notes: Model 2: Adjusted for age, sex, race, BMI, diastolic blood pressure, systolic blood pressure, eGFR, total glucose, total cholesterol, triglycerides, HDL-C, diabetes, current smoking, and history of cardiovascular events.
Abbreviations: ARIC, Atherosclerosis Risk in Communities; BMI, Body mass index; eGFR, estimated glomerular filtration rate; HDL-C, high-density lipoprotein cholesterol; MASLD, Metabolic Dysfunction-associated Steatotic Liver Disease.

cardiovascular disease in individuals with MASLD.[1,7–10,26,27] First, the primary driver of MASLD is overnutrition, which causes the expansion of adipose depots and accumulation of ectopic fat, leading to an immune response. In this context, macrophage infiltration of the visceral adipose tissue compartment creates a proinflammatory state that promotes insulin resistance and contributes to the generation of a systemic inflammatory environment through the secretion of inflammatory cytokines and acute-phase proteins. This persistent, low-grade inflammatory environment fuels cardiovascular disease through endothelial dysfunction, altered vascular tone, enhanced plaque formation, and coagulation.

Table 4 Hazard Ratios of 10-Year Major Adverse Cardiovascular Events and All-Cause Mortality from MASLD Group with FIB-4<2.67

	Model 2 HR (95% CI)	*P* value
Major adverse cardiovascular events		
Leukocytes	1.081 (0.970–1.204)	0.157
Neutrophils	1.110 (0.997–1.235)	0.056
Lymphocytes	0.933 (0.841–1.035)	0.187
Monocytes	1.115 (1.020–1.218)	0.017
C-reactive protein	1.094 (1.007–1.188)	0.034
All-cause mortality		
Leukocytes	1.254 (1.083–1.453)	0.003
Neutrophils	1.296 (1.119–1.501)	0.001
Lymphocytes	0.995 (0.856–1.157)	0.949
Monocytes	1.134 (0.989–1.301)	0.072
C-reactive protein	1.115 (1.009–1.231)	0.032

Notes: Model 2: Adjusted for age, sex, race, BMI, diastolic blood pressure, systolic blood pressure, eGFR, total glucose, total cholesterol, triglycerides, HDL-C, diabetes, current smoking, and history of cardiovascular events.
Abbreviations: ARIC, Atherosclerosis Risk in Communities; BMI, Body mass index; eGFR, estimated glomerular filtration rate; FIB-4, Fibrosis-4 index; HDL-C, high-density lipoprotein cholesterol; MASLD, Metabolic Dysfunction-associated Steatotic Liver Disease.

Table 5 Hazard Ratios of 10-Year MACE and All-Cause Mortality from MASLD Group Not Having History of MACE

	Model 2 HR (95% CI)	*P* value
Major adverse cardiovascular events		
Leukocytes	1.093 (0.956–1.250)	0.192
Neutrophils	1.117 (0.979–1.275)	0.100
Lymphocytes	0.939 (0.826–1.068)	0.338
Monocytes	1.124 (1.017–1.242)	0.021
C-reactive protein	1.107 (1.021–1.201)	0.014
All-cause mortality		
Leukocytes	1.138 (0.918–1.410)	0.239
Neutrophils	1.132 (0.916–1.399)	0.251
Lymphocytes	0.993 (0.807–1.223)	0.949
Monocytes	1.125 (0.943–1.340)	0.190
C-reactive protein	1.090 (0.984–1.208)	0.099

Notes: Model 2: Adjusted for age, sex, race, BMI, diastolic blood pressure, systolic blood pressure, eGFR, total glucose, total cholesterol, triglycerides, HDL-C, diabetes, current smoking, and history of cardiovascular events.
Abbreviations: ARIC, Atherosclerosis Risk in Communities; BMI, Body mass index; eGFR, estimated glomerular filtration rate; HDL-C, high-density lipoprotein cholesterol; MACE, major adverse cardiovascular events; MASLD, Metabolic Dysfunction-associated Steatotic Liver Disease.

Additionally, in the presence of insulin resistance, an imbalance between the influx and efflux of fat leads to the formation of lipotoxic lipids which contribute to cellular stress (mitochondrial dysfunction and endoplasmic reticulum stress) and subsequent inflammation.

Previous studies have extensively discussed the role of monocytes in MASLD.[10] In the early stages of inflammation, metabolic injury to the liver activates different immune cells, including hepatic Kupffer cells, which are liver-resident macrophages that can adopt an inflammatory phenotype and activate other immune cells by releasing inflammatory

cytokines. As inflammation progresses, Kupffer cells are gradually replaced by monocyte-derived macrophages, which can differentiate into proinflammatory macrophages and release cytokines. Our study validated previous findings that increased levels of monocytes were associated with an increased 10-year risk of MACE, which may represent the disease progression.

Our study extended the knowledge on CRP and its association with adverse cardiovascular outcomes. In the MASLD population, levels of CRP are closely related to the risk of MACE in the next 10 years. CRP is a specific protein, and previous studies have demonstrated its association with the severity of histological features of MASLD.[28,29] However, the specific mechanism of CRP elevation is unclear, and it is not certain whether CRP itself is part of the causal pathway of inflammation and cardiovascular events in the MASLD population. A previous Mendelian randomization study showed that CRP is merely a biomarker, not a mediator, of cardiovascular disease risk.[30]

Our study showed that systemic inflammation (measured by monocytes and CRP) was associated with future cardiovascular risk in patients with MASLD and was an independent predictor. Therefore, interventions targeting inflammation may reduce the occurrence of future cardiovascular events in the MASLD population.

Currently, resmetirom, an oral thyroid hormone receptor-β agonist, had been tested in Phase III trials (NCT03900429) and was the first medication approved for use in conjunction with diet and exercise for the treatment of adults with NASH with moderate to advanced liver fibrosis in the USA.[31,32] And some other treatment strategies mainly focus on medication for complications such as type 2 diabetes mellitus.[5,6] Glucose lowering agents, such as glucagon-like peptide-1 receptor agonists and sodium-glucose cotransporter 2 inhibitors, might have different degrees of benefit.[25,33] Based on the exploration of inflammatory cells and their activation pathways, some clinical trials of drugs targeting inflammation in NASH are also undergoing.[34] Monocytes are one direction of exploration, with CC-chemokine ligand (CCL), CC chemokine receptor (CCR), and CD44 being potential therapeutic targets. As MASLD progresses, CCL1, 2, 5, and CD44 can drive the recruitment of monocyte-derived macrophages from the circulation and induce their polarization towards an inflammatory phenotype.[10] Unfortunately, single-agent therapy with cenicriviroc as a CCR2/5 inhibitor, although showing promising results in preclinical and early clinical studies,[35–37] has been terminated early due to a lack of efficacy based on the interim analysis of Part 1 data (NCT03028740).[38] Additionally, research on the neutralization of CD44 has shown some efficacy in animal models,[39] and Phase II clinical trials of combined therapy with CCR2/5 inhibitors and farnesoid X receptor (FXR) agonists are underway (NCT03517540).[10] Further research is needed to understand the mechanisms underlying monocytes and identify promising targets. Furthermore, a possible reason for the failure of previous studies is that improvement in fibrosis was included as the primary outcome. Therefore, we recommend that cardiovascular events could also be included as pre-specified endpoints in future trials.

Limitations

Our study had several Limitations. First, this was a prospective observational study, and residual confounders caused by unknown factors cannot be excluded. Second, some covariates were based on self-reports and the participants must live until visit 2 to be included in this study. Relatedly, hematological biomarkers of inflammation and risk factors for cardiovascular events were recorded at visit 2, without considering changes over time. Third, we excluded participants with high alcohol consumption, but we lacked information on secondary etiologies that may lead to MASLD, such as hepatitis C or hemochromatosis. However, previous studies have suggested a low prevalence of these diseases in our cohort.[40] Finally, since visit 2 of the ARIC did not include liver imaging or liver biopsy, we only use FLI to assess hepatic steatosis. Nonetheless, the current international guidelines recognize the utility of such non-invasive methods in large epidemiologic studies.[5]

Conclusion

In this study, we found that increased levels of monocytes and CRP were associated with an increased 10-year risk of MACE in the MASLD population. Hematological biomarkers of inflammation may help identify MASLD populations at higher risk for cardiovascular events. Interventions targeting inflammation may have the potential to reduce the burden of cardiovascular disease in the MASLD population.

Data Sharing Statement

The data is not publicly available. Interested researchers may contact the NHLBI to apply for data and study materials.

Ethical Statement

The ARIC study has been approved by the Institutional Review Boards (IRB) at all participating institutions: University of North Carolina at Chapel Hill IRB, Wake Forest University IRB, Johns Hopkins University IRB, University of Minnesota IRB and University of Mississippi Medical Center IRB. Written informed consent was obtained from all study participants. All methods were carried out in accordance with the relevant guidelines and regulations for human subject research, in accordance with the Declaration of Helsinki (1989). We have obtained written permission to use the ARIC data from the National Heart, Lung and Blood Institute (NHLBI). We have obtained approval from the Ethics Committee of Guangdong Provincial People's Hospital (reference number: KY-Q-2022-347-01).

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Disclosure

The authors report no potential conflicts of interest in this work.

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