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Neutrophil/Lymphocyte Ratio and All-Cause Mortality in Diabetic Kidney Disease: A Retrospective Cohort Study

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Background: Diabetic kidney disease (DKD) is a significant contributor to the development of end-stage renal disease and cardiovascular disease (CVD), with inflammation being a critical factor in its pathogenesis. The aim of this study is to examine the relationship between the neutrophil-to-lymphocyte ratio (NLR), a new inflammatory marker, and mortality from all causes and CVD in patients with DKD.

Methods: This multicenter, retrospective cohort study utilized data from the China Renal Data System (CRDS) on patients with DKD hospitalized between January 1, 2000, and February 28, 2023. The patients' demographic information, along with their initial clinical and laboratory results, were collected and recorded. Follow-up continued until July 1, 2023, and patients were categorized into two groups based on the median baseline NLR. The Cox proportional hazards regression, Restricted cubic spline (RCS) curves, The Kaplan–Meier curve, Fine–Gray competing risk model, Time-dependent ROC and subgroup analysis were used to analyze the association between all-cause mortality and CVD mortality in patients having DKD with varying NLR.

Results: This study included 11,427 patients who had been clinically diagnosed with DKD. Baseline NLR was associated with C-reactive protein, procalcitonin, high-sensitivity C-reactive protein, plasma D-dimer, cystatin C, creatinine, urea nitrogen, brain natriuretic peptide, and eGFR. We selected the demographic characteristics, differential factors from univariate analysis, and clinically DKD-related laboratory indicators as covariates for Cox analysis. Results indicated that NLR was an independent risk factor for both all-cause and CVD mortality after adjusting for the relevant variables. The risk of all-cause death and CVD death in the high NLR group was 4.688 and 2.141 times higher, respectively, compared to the low NLR group (HR = 4.688, 95% CI 1.153–19.061, P = 0.031; HR = 2.141, 95% CI 1.257–3.644, P = 0.005). However, potential confounding factors and biases, such as unmeasured variables and the influence of treatment interventions, could not be fully accounted for.

Conclusion: NLR can independently predict the risk of all-cause and CVD mortality in patients with DKD. Identifying individuals with a high NLR and providing further intervention could be crucial measures to reduce both all-cause and CVD mortality. However, the results should be interpreted with caution due to the study's limitations.

Keywords: all-cause death, cardiovascular disease, diabetic kidney disease, neutrophil-to-lymphocyte ratio, renal function

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Introduction

The prevalence of diabetes mellitus (DM) has surged worldwide, with the number of individuals with diabetes projected to reach 643 million by 2030.[1] Diabetic kidney disease (DKD) is a prevalent chronic microvascular complication of DM.[2,3] Studies indicate that approximately 30% of patients with type 1 diabetes mellitus (T1DM) and 40% of type 2 diabetes mellitus (T2DM) patients develop DKD.[4,5] In a large cohort study of half a million people monitored for an average of 8 years, all-cause mortality was twice as high in those with early-stage DKD compared to those with "diabetes without chronic kidney disease (CKD)" or "CKD without diabetes."[6] Additionally, some studies have shown that cardiovascular disease (CVD) is the leading cause of death among patients with DKD.[7–10] Consequently, introducing new biomarkers to predict future risks of all-cause and CVD mortality in the DKD population is crucial to reduce mortality rates.

Inflammation and the immune response are vital in the development and progression of DKD.[11–13] For example, patients with early-stage DKD exhibit greater infiltration of immune cells, including T cells and B cells, in kidney tissue compared to those with non-diabetic kidney diseases.[14] Additionally, the expression of TNFRSF21, a circulating inflammatory protein and a KRIS marker linked to end-stage renal disease (ESRD), is elevated in the CD14+ monocyte subset of patients with DKD.[15] This suggests that immune cells may impact the prognosis of patients with DKD.

The neutrophil-to-lymphocyte ratio (NLR) is a simple and cost-effective laboratory indicator obtained from routine blood tests. It reflects both the adaptive immune response mediated by lymphocytes and the innate immune response mediated by neutrophils, making it an increasingly recognized inflammatory biomarker.[16] Numerous studies have demonstrated NLR's predictive value for prognosis in CVD, infectious diseases, cancer, and more, highlighting its potential clinical utility.[17–19] For example, elevated NLRs are significantly associated with higher risks of all-cause mortality and cardiovascular mortality among the elderly.[20] Additionally, research has shown that higher NLRs are linked to a greater prevalence of renal complications in patients with DM.[21] While most existing literature focuses on the relationship between NLR and renal function/prognosis in patients with CKD, there is a lack of multicenter studies with large sample sizes examining the association between NLR and all-cause mortality, particularly in patients with DKD.[22,23] Therefore, the aim of our study is to investigate the relationship between NLR and the risk of cardiovascular and all-cause mortality in the patient population with DKD. This research could provide valuable insights for stratifying patients with DKD and guiding their management and treatment.

Methods

Study Design and Data Sources

The study sample was sourced from the China Renal Data System (CRDS).[24] Established in December 2018 and led by the National Clinical Research Center for Kidney Disease and the Chronic Disease Center of The Chinese Center for Disease Control and Prevention, the CRDS is a national multicenter network of regional medical centers. The registry includes demographic and clinical data on patients admitted to participating centers, with data exported from its proprietary hospital information system since 2000. As of August 1, 2023, the CRDS database contains information from over 8 million patients across 24 medical centers. The exported data were clinical data that had been desensitized, ensuring patient privacy was not compromised.

Study Population

Patients with DKD admitted to the hospital between January 1, 2000, and February 28, 2023, were selected from the CRDS database, which had been cleaned, standardized, anonymized, and merged. According to the 2012 KDIGO Clinical Practice Guidelines, DKD was defined as a urinary albumin-to-creatinine ratio (UACR) of $\geq$ 30 mg/g or an estimated glomerular filtration rate (eGFR) of < 60 mL/min/1.73m², with no other primary or secondary kidney diseases.[25] We specified exclusion criteria to take into account the purpose of our study and the effect of certain diseases and drugs on the results of our analysis (see the Supplementary Material for details). Patients meeting any of the following criteria were excluded: (1) under 18 years old; (2) incomplete clinical data; (3) follow-up time less than 6 months; (4) diagnosed with ESKD or receiving renal replacement therapy before discharge; (5) acute or chronic

infection, acute coronary disease, heart failure, cancer, surgery, or thyroid disease; (6) hematological diseases affecting blood cell count (eg, leukemia), kidney injury from other factors (such as nephrotic syndrome), or autoimmune diseases; (7) taking drugs affecting lymphocyte and neutrophil counts (such as steroids) or had abnormal neutrophil and lymphocyte values. The sample selection flow chart is shown in Figure 1.

Variables and Definition

We collected demographic data, which encompassed details such as gender, birthdate, height, weight, as well as systolic and diastolic blood pressure. We also extracted laboratory test results, including A/G:Albumin/globulin ratio; ALP: alkaline phosphatase; ALT: Alanine Aminotransferase; AST: Aspartate Transaminase; B2MG: beta 2-microglobulin; BNP: brain natriuretic peptide; BUN: blood urea nitrogen; Scr: serum creatinine; CRP: C-reactive protein; Cysc: cystatin C; dbil: direct bilirubin; dd2: D-Dimer; fib: fibrinogen; Hb: hemoglobin; hcy: homocysteine; hdl-c: high density lipoprotein cholesterol; hscrp: high-sensitivity C-reactive protein; IBIL: Indirect Bilirubin; K: potassium ion; LDL: low density lipoprotein cholesterol; Na: Sodium ion; P: Phosphorus; PCT: procalcitonin; PLT: platelet count; PTH: parathyroid hormone; RBC: Red blood cells; TBIL: total bilirubin; TC: total cholesterol; TG: triglyceride; TP: total protein; UA: uric acid; UACR: urinary albumin to creatinine ratio; WBC: white blood cells. Baseline information referred to the measurements obtained from the final clinical test conducted before hospital discharge for patients diagnosed with diabetic kidney disease upon initial admission. All covariates were chosen based on their biological significance or their relevance in previously published studies, which were considered clinically significant.

The eGFR was determined using the 2009 Chronic Kidney Disease Epidemiology Collaboration equation.[26] The NLR was calculated by dividing the absolute neutrophil count (10^9/L) by the absolute lymphocyte count (10^9/L), while the body mass index (BMI) was calculated as weight divided by height squared (kg/m^2). End-stage kidney disease (ESKD) was defined as either maintenance dialysis, kidney transplantation, or an eGFR < 15 mL/min/1.73m^2. CVD death was characterized as death resulting from coronary events, arrhythmia, sudden cardiac death, congestive heart failure, or cerebrovascular events.

Follow-Up and Study Outcomes

NLR was computed based on the findings of the final routine blood test conducted before discharge. Patients were categorized into two groups determined by the median NLR (M) of the study population: (1) the low NLR group (≤ M); (2) the high NLR group (> M). The primary endpoints of the study were all-cause mortality and CVD mortality, and all participants underwent follow-up. The follow-up duration extended beyond 6 months from the initial hospital discharge diagnosis of DKD until the occurrence of any outcome, loss to follow-up, or February 28, 2023, whichever came first.

Statistical Analysis

The included data were analyzed using the R software version 4.3.1, with statistical significance set at $P < 0.05$ (two-sided). In cases of limited missing data, we employed multiple imputation based on chained equations to address missing values for variables. Continuous variables with a normal distribution are reported as mean ± standard deviation and were compared between groups using t-tests. Variables with a skewed distribution were presented as median (upper quartile, lower quartile) and were compared between groups using nonparametric tests. Count data were expressed as frequencies (percentages), and the chi-square test was utilized for group comparisons.

The association between baseline variables and the NLR was evaluated using Spearman correlation analysis. The cumulative survival rate of each group was estimated using the Kaplan–Meier method, and survival curves were plotted, with the Log rank test employed to compare survival differences between groups. Cox regression analysis was utilized to determine the predictive value of the NLR for clinical outcomes, specifically all-cause mortality and CVD mortality. We utilized a competing risk model to analyze multiple endpoints, reporting hazard ratios (HRs) along with their corresponding 95% confidence intervals (CIs). To visualize the relationship between NLR and HR, a restricted cubic spline (RCS) model was fitted at four nodes (P5, P35, P65, and P95).[27] Additionally, a time-dependent ROC curve was used to assess the predictive capability of the NLR for all-cause mortality in patients with DKD.

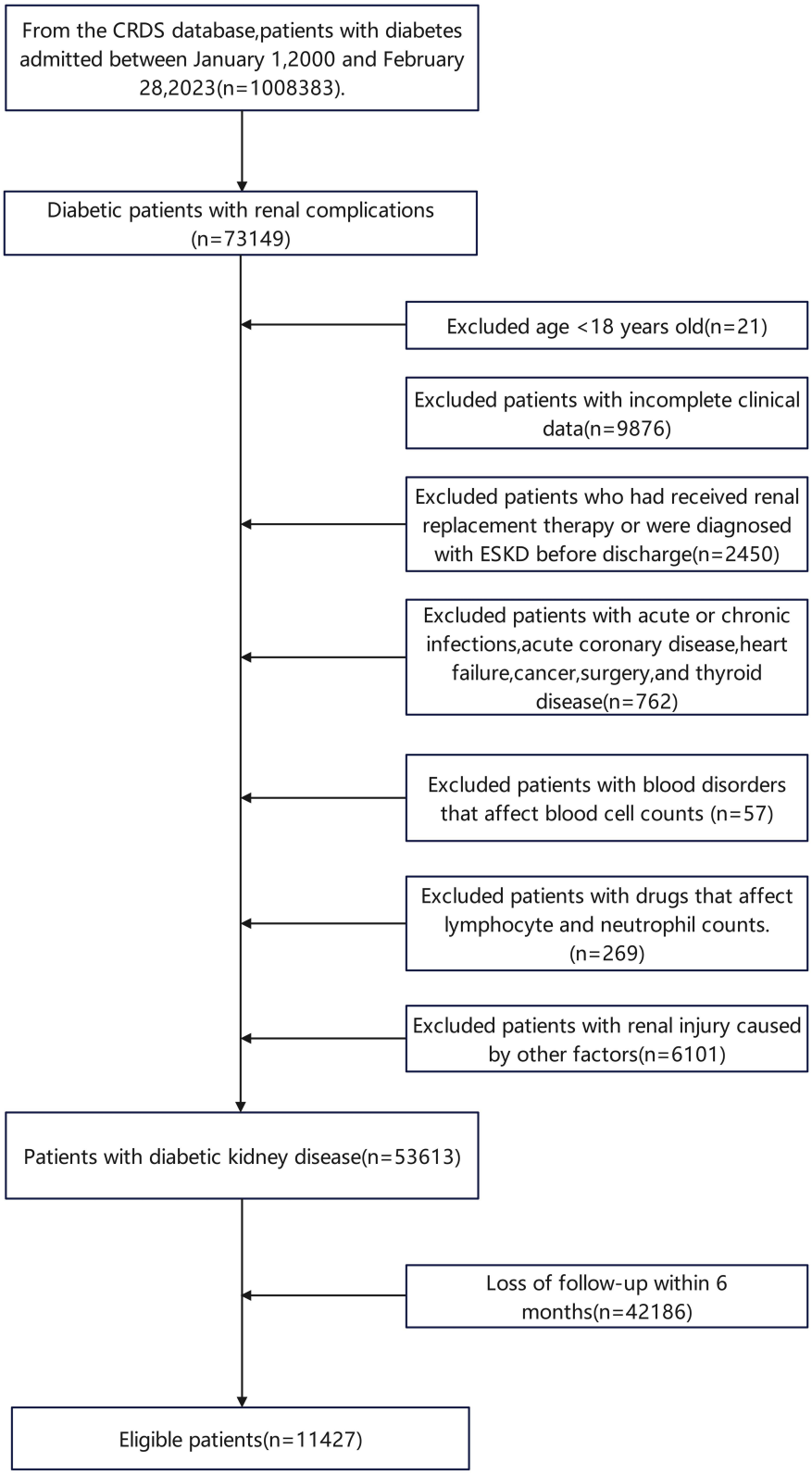

Figure 1 Flowchart of sample selection from the CRDS.

Ethics Approval

Ethics Approval Statement

The study protocol received approval from the Medical Ethics Committee of Nanfang Hospital, Southern Medical University (NFEC-2019-213), as well as from the China Office of Human Genetic Resources for Data Preservation Application (2021-BC0037).

Patient Consent Statement

Patient consent was not required due to the retrospective design of the study. The research was carried out in compliance with the Declaration of Helsinki and adhered to the Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) guidelines.[28]

Results

This study included 11,427 patients who had been clinically diagnosed with DKD, comprising 6734 males (58.93%) and 4693 females (41.07%). The median age was 63 years (interquartile range: 54.00, 71.00), with a follow-up period of 62 months (interquartile range: 44, 83).

The Baseline Characteristics of the Patients Were Grouped According to the Median Baseline NLR

The study cohort had a median NLR of 3.17 (interquartile range: 2.15, 4.93). Table 1 presents the baseline characteristics of the 11,427 patients with DKD, stratified by the median NLR. These patients were subsequently divided into two groups based on their NLR: the low NLR group (NLR ≤ 3.17, n = 5714) and the high NLR group (NLR > 3.17, n = 5714). Compared to the low NLR group, individuals in the high NLR group were older, had a higher proportion of males, and showed associations with elevated levels of systolic blood pressure, alkaline phosphatase, aspartate aminotransferase, β2-microglobulin, brain natriuretic peptide, urea nitrogen, creatinine, C-reactive protein, cystatin-C, plasma D-dimer, ferritin, plasma fibrinogen, γ-glutamyltransferase, homocysteine, hypersensitive C-reactive protein, potassium ion, inorganic phosphorus, procalcitonin, platelet count, parathyroid hormone, uric acid, urinary albumin/creatinine ratio, and white blood cell count (all $P < 0.05$). Conversely, BMI, the albumin/globulin ratio, alanine aminotransferase, calcium ion, iron ion, hemoglobin, indirect bilirubin, low-density lipoprotein cholesterol, sodium ion, red blood cell count, total cholesterol, triglyceride level, and total protein were observed to be lower in the low NLR group (all $P < 0.05$). Additionally, individuals in the high NLR group had a higher incidence of all-cause mortality (n = 1394) and CVD mortality (n = 348) compared to those in the low NLR group.

Correlations Between NLR and Other Biochemical Parameters

In patients diagnosed with DKD, we used Spearman correlation analysis to explore the connections between the NLR and various biochemical and inflammatory parameters (see Table 2). Notably, the NLR exhibited significant positive correlations with CRP ($r = 0.471$, $P < 0.001$), procalcitonin ($r = 0.453$, $P < 0.001$), white blood cell count ($r = 0.395$, $P < 0.001$), high-sensitivity C-reactive protein ($r = 0.392$, $P < 0.001$), D-dimer ($r = 0.356$, $P < 0.001$), cystatin C ($r = 0.352$, $P < 0.001$), creatinine ($r = 0.343$, $P < 0.001$), urea nitrogen ($r = 0.333$, $P < 0.001$), and brain natriuretic peptide ($r = 0.313$, $P < 0.001$). Conversely, eGFR exhibited a significant negative correlation ($r = -0.349$, $P < 0.001$).

Risk Factors for All-Cause Death and CVD Death in Patients with DKD

After accounting for various confounding factors, as illustrated in Figure 2, Cox regression analysis demonstrated that NLR independently predicted both all-cause mortality and CVD mortality. In the high NLR group, there was a notable increase in the risk of all-cause mortality by 4.688 times (HR = 4.688, 95% CI 1.153–19.061, P = 0.031) and CVD mortality by 2.141 times (HR = 2.141, 95% CI 1.257–3.644, P = 0.005). Furthermore, subgroup analyses were performed based on age, gender, CRP level, and eGFR. Our findings revealed a significant correlation between NLR and all-cause mortality in DKD patients ($P < 0.05$). While there was some variation in specific HR values among subgroups, the relationship between NLR and all-cause mortality remained consistent across all strata ($P > 0.05$ for all interactions),

Table 1 Baseline Characteristics of Participants According to NLR.

Variable	low NLR group (n=5714)	High NLR group (n=5713)	*P-value*
male	3264(56.9)	3470(61.0)	<0.001
female	2477(43.2)	2216(39.0)	<0.001
age	61.09 (12.29)	63.95 (11.74)	<0.001
BMI	24.49 (3.73)	23.47 (3.72)	<0.001
SBP	140.44 (21.48)	144.34 (25.13)	<0.001
DBP	81.57 (12.73)	81.33 (13.75)	0.65
A/G	1.29 (0.33)	1.16 (0.32)	<0.001
ALP	77.00 [62.00, 98.00]	88.00 [69.00, 115.00]	<0.001
25-hydroxyvitamin D	12.96 [8.66, 18.16]	11.50 [6.85, 18.06]	0.12
ALT	17.00 [12.00, 25.58]	16.00 [10.80, 25.00]	<0.001
AST	19.00 [15.00, 26.00]	20.00 [15.00, 28.50]	0.006
B2MG	6.09 [3.14, 14.83]	13.56 [5.59, 20.89]	<0.001
BNP	89.10 [36.23, 324.60]	287.50 [88.00, 1019.34]	<0.001
BUN	10.85 (7.04)	15.27 (8.90)	<0.001
BUN/Scr	40.78 (37.55)	40.98 (37.28)	0.886
Ca	1.13 (0.32)	1.08 (0.17)	0.006
Scr	124.00 [80.00, 266.00]	248.60 [123.40, 516.00]	<0.001
CRP	4.00 [1.53, 6.35]	9.80 [4.58, 47.81]	<0.001
high-sensitivity troponin	0.01 [0.00, 0.03]	0.01 [0.00, 0.07]	0.143
Cysc	1.67 [1.08, 2.96]	2.89 [1.69, 4.66]	<0.001
dbil	2.20 [1.40, 3.40]	2.30 [1.30, 3.60]	0.588
dd2	0.52 [0.27, 1.10]	0.98 [0.50, 2.13]	<0.001
Fe	10.90 [7.80, 14.80]	9.20 [6.00, 13.05]	<0.001
ferritin	336.19 (374.06)	403.62 (413.87)	0.002
fib	3.87 (1.21)	4.53 (1.49)	<0.001
gamma-glutamyltransferase	25.00 [17.00, 41.00]	29.00 [18.80, 55.00]	<0.001
Hb	113.15 (24.98)	100.40 (25.06)	<0.001
Total hemoglobin A1c	9.11 (1.79)	9.12 (1.93)	0.98
hcy	17.68 (9.45)	20.36 (9.65)	<0.001
hdl-c	1.14 (0.39)	1.13 (0.42)	0.078
hscrp	2.42 [0.81, 5.20]	5.00 [2.40, 24.90]	<0.001
IBIL	5.50 [3.40, 8.80]	5.40 [3.10, 9.00]	0.09
K	4.25 (0.65)	4.39 (0.93)	<0.001
LDL	3.04 (1.31)	2.88 (1.33)	<0.001
Na	139.66 (3.89)	137.91 (5.11)	<0.001
P	1.25 [1.09, 1.46]	1.32 [1.08, 1.65]	<0.001
PCT	0.89 (10.16)	3.41 (15.35)	<0.001
PLT	224.19 (79.07)	235.47 (100.33)	<0.001
PTH	73.80 [39.60, 175.70]	154.37 [70.17, 282.00]	<0.001
RBC	3.92 (0.87)	3.52 (0.86)	<0.001
TBIL	8.10 [5.40, 11.80]	8.00 [5.30, 12.00]	0.983
TC	5.11 (1.77)	4.77 (1.69)	<0.001
TG	1.66 [1.16, 2.46]	1.47 [1.04, 2.19]	<0.001
TP	64.56 (9.18)	62.93 (9.02)	<0.001
UA	397.55 (122.58)	420.79 (142.84)	<0.001
UACR:	1453.53 (3284.04)	2428.29 (3473.80)	<0.001
WBC	6.73 (2.07)	8.79 (3.98)	<0.001
all-cause mortality			
0	4845	4292	
1	896	1394	<0.001

(*Continued*)

Table 1 (Continued).

Variable	low NLR group (n=5714)	High NLR group (n=5713)	*P-value*
CVD mortality			
0	4826 (84.5)	4313 (75.5)	<0.001
1	221 (3.9)	348 (6.1)	
2	667 (11.7)	1052 (18.4)	

Notes: Continuous variables were expressed as mean (standard deviation) or median (25th percentile −75th percentile). Categorical variables were expressed as number (percent).

Abbreviations: BMI: the body mass index; SBP, systolic blood pressure; DBP, Diastolic Blood Pressure; A/G, Albumin/globulin ratio; ALP, alkaline phosphatas; ALT, Alanine Aminotransferase; AST, Aspartate Transaminase; B2MG, beta 2-microglobulin; BNP, brain natriuretic peptide; BUN, blood urea nitrogen; Scr, serum creatinine; CRP, C-reactive protein; Cysc, cystatin C; dbil, direct bilirubin; dd2, D-Dimer; fib, fibrinogen; Hb, hemoglobin; hcy, homocysteine; hdl-c, high density lipoprotein cholesterol; hscrp, high-sensitivity C-reactive protein; IBIL, Indirect Bilirubin; K, potassium ion; LDL, low density lipoprotein cholesterin; Na, Sodium ion; P, Phosphorus; PCT, procalcitonin; PLT, platelet count; PTH, parathyroid hormone; RBC, Red blood cells; TBIL, total bilirubin; TC, total cholesterol; TG, triglyceride; TP, total protein; UA, uric aci; UACR, urinary albumin to creatinine ratio; WBC, white blood cells.

Table 2 Spearman Correlation Analysis Between NLR and Baseline Variables

Variable	NLR	
	r^a	P-value
BNP	0.3134	P<0.001
BUN	0.3334	P<0.001
Scr	0.3425	P<0.001
CRP	0.4707	P<0.001
Cysc	0.352	P<0.001
dd2	0.3559	P<0.001
hscrp	0.3917	P<0.001
PCT	0.453	P<0.001
eGFR	−0.3486	P<0.001

Note: [a]r represents the Spearman correlation coefficient r value.

Abbreviations: NLR, neutrophil-to-lymphocyte ratio; BNP, brain natriuretic peptide;BUN, blood urea nitrogen; Scr, serum creatinine; CRP, C-reactive protein; Cysc, cystatin C; dd2, D-Dimer; hscrp, high-sensitivity C-reactive protein; PCT, procalcitonin; eGFR, estimated glomerular filtration rate.

indicating no significant dependence of age, sex, CRP level, or eGFR among patients (Table 3). Figure 3 illustrates a substantial rise in the risk of all-cause mortality with increasing NLR values. However, beyond a value of 10, although the risk continues to increase, it does so at a slower pace. Notably, a nonlinear relationship is evident between the NLR and all-cause mortality (nonlinear effect size = 118.84, P < 0.001).

Association of NLR with All-Cause and CVD Mortality in Patients with DKD

At the end of the follow-up period, the study included a total of 11,427 patients, of whom 2289 (20.03%) had passed away. Among these fatalities, 570 (24.90%) were attributed to CVD, while non-CVD causes accounted for 1720 deaths (75.14%). Kaplan-Meier survival analysis (depicted in Figure 4) revealed a significantly higher overall survival rate in the low NLR subgroup compared to the high NLR subgroup (log-rank X2=380, P<0.001). Even after adjusting for competing risk events (illustrated in Figure 5), the high NLR group still demonstrated an increased risk of all-cause mortality (P < 0.001). Considering all-cause mortality as the outcome, the cumulative survival rates at 1 year, 3 years, and 5 years were 97.6%, 91.5%, and 86.9%, respectively, in the low NLR subgroup, whereas they were 94.7%, 83.7%, and 77.6%, respectively, in the high NLR subgroup.

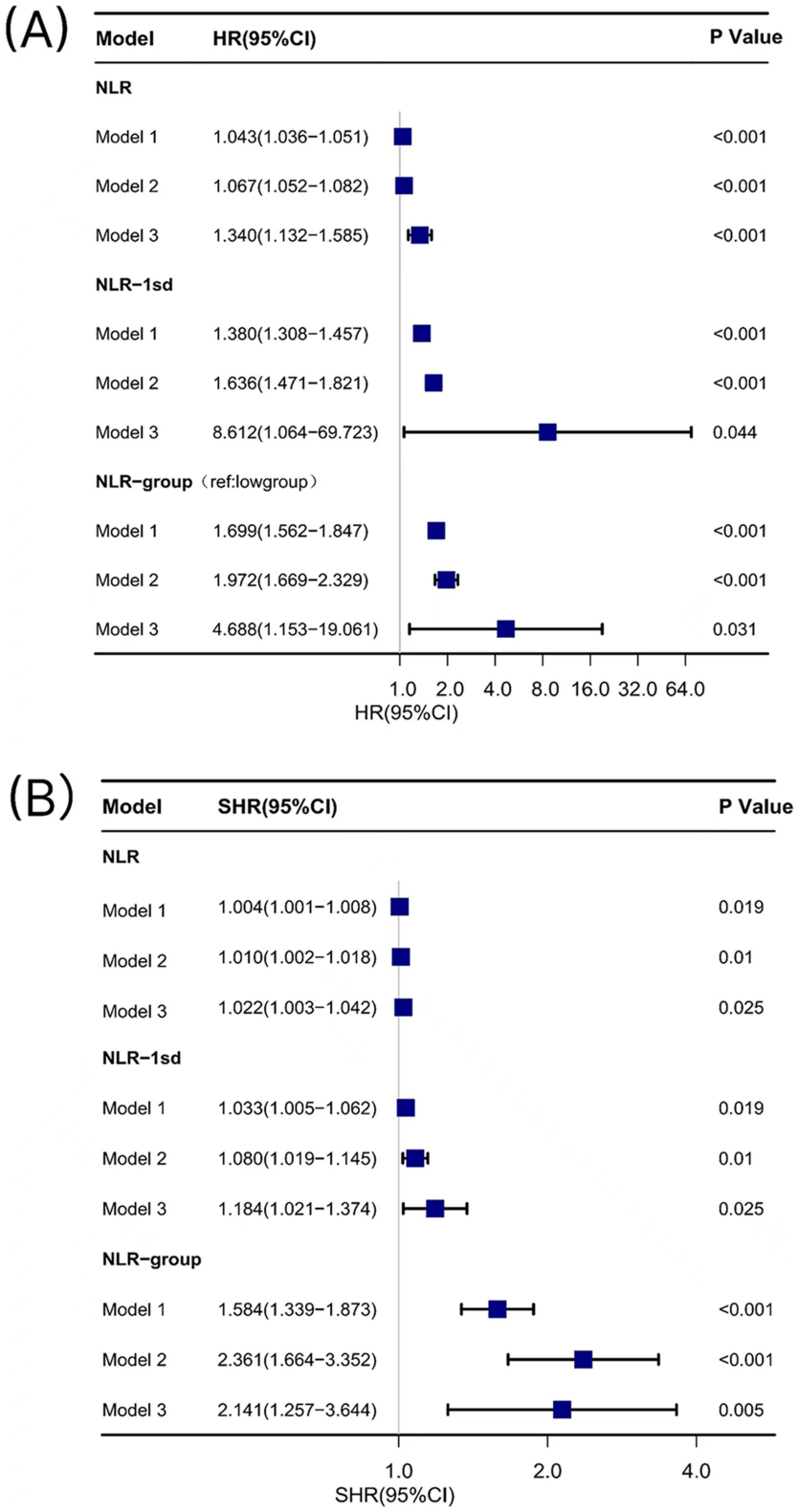

Figure 2 Forest plot depicting Cox regression analysis results illustrating the relationship between baseline NLR and all-cause mortality (**A**) and CVD mortality (**B**). **Model 1**: unadjusted; **Model 2**: adjusted for age, sex, systolic blood pressure, diastolic blood pressure, and BMI; **Model 3**: The levels of leukocyte ratio, alkaline phosphatase, total cholesterol, alanine aminotransferase, aspartate aminotransferase, urea nitrogen, creatinine, C-reactive protein, cystatin C, direct bilirubin, plasma fibrinogen, γ-glutamine, white blood cell, high density lipoprotein, indirect bilirubin, potassium ion, low density lipoprotein, sodium ion, inorganic phosphorus, platelet count, red blood cell, total bilirubin, total were corrected; Cholesterol, triglyceride, total protein, uric acid, eGFR, and CVD death were further adjusted for WBC ratio, alkaline phosphatase, hydroxyvitamin D, alanine aminotransferase, aspartate aminotransferase, total cholesterol, total protein, and uric acid. **NLR-1sd**: Standardization of NLR continuous variables was performed before entering them into the analysis model. **NLR-group**: The median NLR was utilized as the threshold point and converted into a binary variable before inclusion in the analysis model.

Table 3 Subgroup Analysis of Neutrophil-to-Lymphocyte Ratio (NLR) and All-Cause Mortality

Subgroup	P for interaction	HR(95% CI)	P-value
Age(years) ≥60	0.00123	1.491(1.34–1.65)	<0.001
Age(years) <60		1.995(1.73–2.30)	<0.001
Sex-Male	0.403	1.748(1.57–1.95)	<0.001
Sex-Female		1.624(1.42–1.86)	<0.001
CRP(mg/L) ≥3.2	0.141	1.861(1.56–2.23)	<0.001
CRP(mg/L) <3.2		1.458(1.11–1.92)	0.006
eGFR(mL/min/1.73m^2) ≥60	0.557	1.676(1.53–1.83)	<0.001
eGFR(mL/min/1.73m^2) <60		1.545(1.20–2.00)	<0.001

Figure 6 depicts the time-dependent receiver operating characteristic (ROC) curve of NLR for predicting mortality. The area under the curve (AUC) for predicting all-cause mortality at 3, 5, and 10 years in patients with DKD was 0.62 (0.56–0.69), 0.61 (0.57–0.65) and 0.62 (0.58–0.67), respectively.

Discussion

The pathogenesis of DKD remains partially understood. However, DKD is characterized by progressive renal function decline and represents a significant contributor to mortality among individuals with diabetes.[29,30] Peripheral white blood cells, including neutrophils and lymphocytes, have been implicated in renal inflammation in patients with DKD and are commonly utilized as readily available inflammatory indicators in clinical practice.[31,32] Moreover, several studies have identified an elevated neutrophil count as a dependable and independent risk factor for CKD progression in patients with diabetes.[33,34] Conversely, T lymphocytes have been shown to confer protection against diabetes and atherosclerosis.[35] In recent years, the novel inflammatory biomarker NLR, reflecting alterations in neutrophils and lymphocytes concurrently, has gained attention. Studies have demonstrated that NLR is stable in a much more superior manner compared to individual levels of neutrophils, lymphocytes, and white blood cells.[21] Therefore, evaluating the association between NLR and all-cause mortality in patients with DKD is crucial.

This retrospective cohort study, notable for its sizable sample size, marks the first instance where the NLR is identified as a risk factor for both all-cause and cardiovascular mortality in patients with DKD. NLR emerges as an independent, dependable, and practical biomarker for predicting both all-cause and cardiovascular mortality in patients with DKD.

Previous research has shown a correlation between NLR and mortality; however, the findings have been inconsistent. For example, some studies have found limited predictive ability of the NLR for cardiovascular mortality following acute coronary syndrome in patients with CKD.[36] Moreover, investigations involving patients with CKD stages 1–4 have not consistently identified a significant association between abnormal NLR levels and the risk of CVD or all-cause mortality.[23] In contrast, our study demonstrated that NLR serves as a stable predictor of CVD death and all-cause mortality over a 10-year period in patients with DKD, with similar predictive value for both outcomes. These discrepancies may stem from differences in the study populations. Our focus was specifically on individuals with diabetes-related chronic kidney disease, unlike studies that included all individuals with CKD regardless of etiology. Conducted within a DKD-diagnosed cohort, our study boasted a large sample size and long follow-up duration (11,427 individuals, 5.17 years), lending greater credibility to our conclusions. By further subgrouping patients based on the specific etiology of chronic kidney disease, our findings underscore the substantial involvement of immune cells in the pathological progression of diabetic kidney disease. Additionally, prior research has indicated that higher NLR is linked to increased incidence of CVD and DKD among diabetic adults, and these findings are consistent with our own.[21] Studies in Asian populations have also suggested that elevated NLR levels may play a significant role in DKD progression, aligning partially with our results.[37,38] Given that a majority of patients with DKD succumb to CVD before reaching ESRD, predicting cardiovascular mortality in patients with DKD not yet on dialysis therapy is of particular importance.[39]

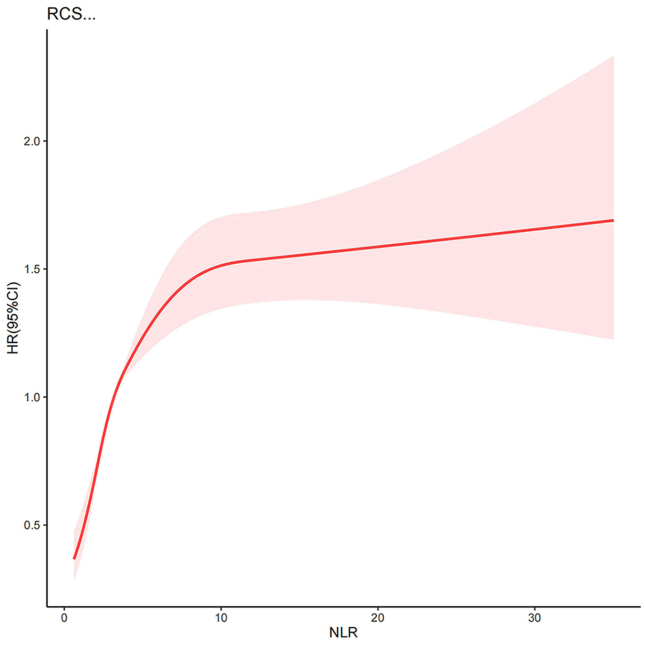

Figure 3 Restricted cubic spline (RCS) curves of NLR in model 1 (full model).

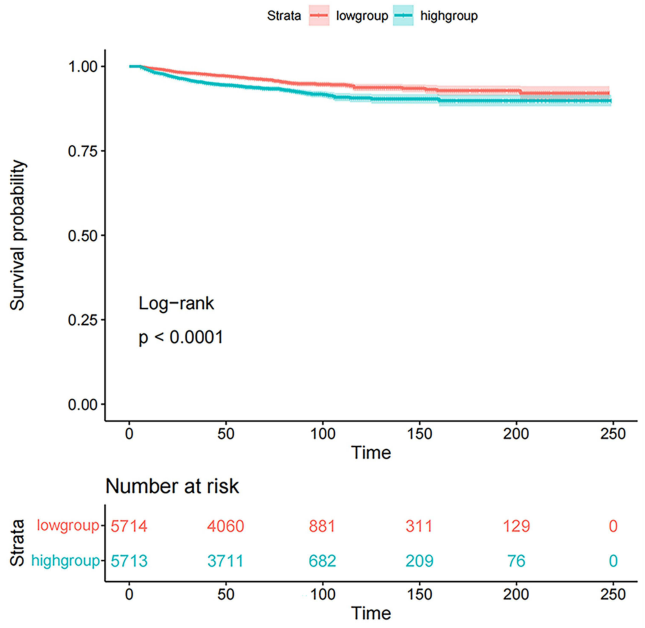

Figure 4 The Kaplan–Meier curve for the low NLR and high NLR group.

Our study unveiled a notable positive correlation between the NLR and inflammatory markers, including procalcitonin, C-reactive protein, and high-sensitivity C-reactive protein. Previous research has indicated that intracellular damage-associated molecular patterns and inflammatory cytokines can foster a proinflammatory milieu, potentially exacerbating DKD severity and mortality.[40] Thus, we hypothesize that the heightened inflammatory status observed in patients with high NLR may contribute significantly to their elevated risk of all-cause mortality compared to those with low NLR.

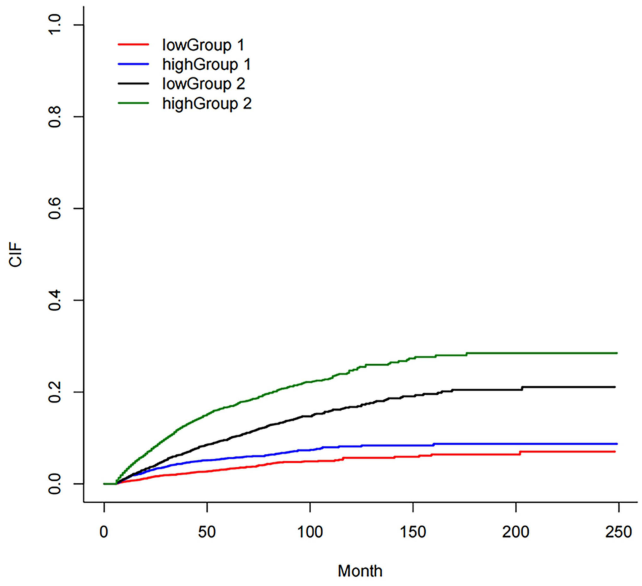

Figure 5 Fine-Gray competition competing risk curve.

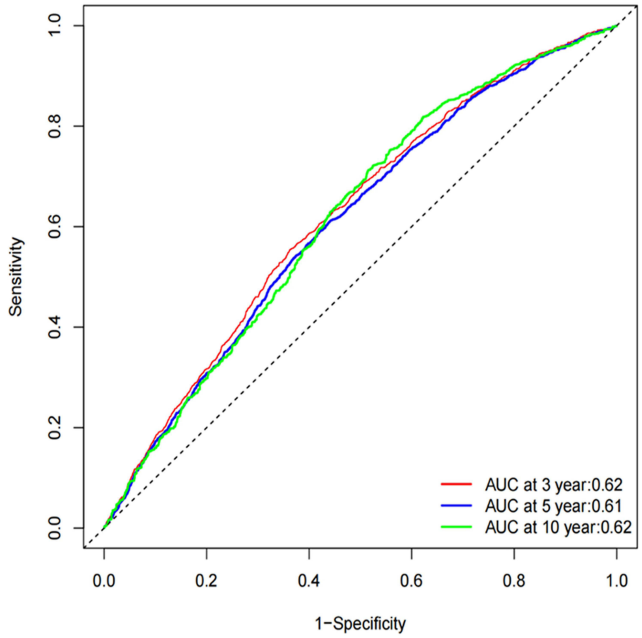

Figure 6 Time-dependent ROC curve of NLR for prediction of all-cause mortality.

However, the NLR, which serves as an alternative inflammation marker, may be affected by other co-existing inflammatory or pathological states in patients with DKD, thereby affecting the accuracy of assessment. In this study, in order to avoid the influence of these factors on the results, we excluded a variety of related diseases, including acute or chronic infections and acute coronary artery disease, etc., to ensure the reliability of the results. However, in clinical practice, because patients often have comorbidities that affect blood cell counts, this limits the application of NLR. Furthermore, at present, different research institutions and laboratories have not yet reached a unified standard to define the normal reference value range of NLR.[41,42] This phenomenon may be due to differences in sample size, population characteristics, and inconsistent detection methods, which further exploration is needed in the future.

Furthermore, our findings revealed a positive correlation between plasma D-dimer levels and NLR, a relationship not previously reported in patients with DKD. However, a case-control study has suggested an association between coagulation imbalance and arterial stiffness and endothelial dysfunction in patients suffering from renal failure.[43] Given that plasma D-dimer levels reflect activation of coagulation and fibrinolytic systems and are elevated in patients with CKD, we speculate that within the chronic inflammatory microenvironment, neutrophils may exacerbate endothelial injury, potentially leading to microvascular thrombosis and worsening renal damage in patients with diabetes.[44–46] This could help elucidate the link between diabetic nephropathy and CVD mortality. Moreover, our study identified a negative correlation between the NLR and renal function indicators such as eGFR, and a positive correlation between the NLR and both blood urea nitrogen and serum creatinine levels. Numerous studies focusing on renal insufficiency have highlighted NLR as an independent prognostic factor for renal function decline. For instance, in patients with IgA nephropathy, a higher NLR was associated with deteriorating renal function.[47] Similarly, in adults with type 2 diabetes, a higher baseline NLR was predictive of declining renal function or proteinuria progression during follow-up.[48] Additionally, among elderly individuals diagnosed with metabolic syndrome, a higher NLR ($\geq$ 3.83) was associated with decreased eGFR.[49] These findings suggest the potential utility of the NLR as a predictive marker for assessing renal function.

While this study offers a comprehensive analysis of real-world data from a large and diverse sample, several limitations warrant acknowledgment. Firstly, although our multicenter data predominantly originate from reputable tertiary hospitals in prefecture-level cities, the absence of data from Northwest China and remote areas may introduce selection bias. Secondly, despite our findings showing an association between the NLR and mortality in patients with DKD, these associations cannot be directly established as causality due to the retrospective design of the study. Lastly, despite employing multivariate Cox regression analysis and adjusting for potential confounders, the presence of unaccounted confounding factors cannot be ruled out entirely.

Conclusions

In Chinese patients with DKD who were not undergoing renal replacement therapy at baseline, high NLR showed a significant association with long-term risks of both all-cause and CVD mortality. These results indicate that NLR can autonomously predict the risk of all-cause and CVD mortality in patients with DKD. Therefore, individuals with high NLR could be crucial targets for additional interventions aimed at reducing both all-cause and CVD mortality.

Abbreviations

DM, diabetes mellitus; DKD, Diabetic kidney disease; T1DM, type 1 diabetes mellitus; T2DM, type 2 diabetes mellitus; CVD, cardiovascular disease; ESRD, end-stage renal disease; NLR, neutrophil-to-lymphocyte ratio; eGFR, estimated glomerular filtration rate;BMI, the body mass index; SBP, systolic blood pressure; DBP, Diastolic Blood Pressure; A/G:Albumin/globulin ratio; ALP, alkaline phosphatas; ALT, Alanine Aminotransferase; AST, Aspartate Transaminase; B2MG, beta 2-microglobulin; BNP, brain natriuretic peptide; BUN, blood urea nitrogen; Scr, serum creatinine; CRP:C-reactive protein; Cysc, cystatin C; dbil:direct bilirubin; dd2, D-Dimer; fib:fibrinogen; Hb, hemoglobin; hcy, homocysteine; hdl-c, high density lipoprotein cholesterol; hscrp, high-sensitivity C-reactive protein; IBIL, Indirect Bilirubin; K, potassium ion; LDL, low density lipoprotein cholesterin; Na, Sodium ion; P, Phosphorus; PCT, procalcitonin; PLT, platelet count; PTH, parathyroid hormone; RBC, Red blood cells; TBIL, total bilirubin; TC, total cholesterol; TG, triglyceride; TP, total protein; UA, uric aci; UACR, urinary albumin to creatinine ratio; WBC: white blood cells.

Data Sharing Statement

All data generated or analysed during this study are included in this article. Further enquiries can be directed to the corresponding author.

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Author Contributions

All authors made a significant contribution to the work reported, whether that is in the conception, study design, execution, acquisition of data, analysis and interpretation, or in all these areas; took part in drafting, revising or critically reviewing the article; gave final approval of the version to be published; have agreed on the journal to which the article has been submitted; and agree to be accountable for all aspects of the work.

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Disclosure

The authors declare that they have no competing interests.

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