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Pretreatment Neutrophil-to-Lymphocyte Ratio as a Risk Marker for Brain Metastasis in Patients with Stage I–III Non-Small Cell Lung Cancer Receiving Definitive Thoracic Radiotherapy

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Abstract

Background

Brain metastasis (BM) represents a major pattern of distant failure in non-small cell lung cancer (NSCLC) following definitive thoracic radiotherapy (RT). Systemic inflammation plays a critical role in tumor progression and the metastatic cascade. This study aimed to evaluate the prognostic value of the baseline neutrophil-to-lymphocyte ratio (NLR) for predicting the occurrence, cumulative incidence, and temporal dynamics of brain metastasis in patients with Stage I–III NSCLC receiving definitive thoracic RT.

Methods

A total of 138 patients with pathologically confirmed Stage I–III NSCLC who received definitive-intent thoracic RT (60–66 Gy) between 2014 and 2024 were retrospectively analyzed. Pre-treatment hematological parameters were obtained within two weeks prior to any oncological intervention. Time-dependent Receiver Operating Characteristic (ROC) curve analysis evaluated the dynamic predictive performance of baseline NLR. Independent predictors of brain failure were identified using cause-specific Cox proportional hazards regression, accounting for death as a competing risk. A pre-specified sensitivity analysis was restricted to Stage III patients (n = 108) with mandatory baseline and routine follow-up brain MRI.

Results

Over a median follow-up of 28.0 months, 40 patients (29.0%) developed BM. Median baseline NLR was significantly higher in patients who developed BM compared to those who did not (3.21 vs. 2.22; $p < 0.001$). Time-dependent ROC analysis at 28 months yielded an Area Under the Curve (AUC) of 0.79, with an optimal NLR threshold of 2.26 (sensitivity: 91.7%, specificity: 54.6%). Multivariable cause-specific Cox regression demonstrated that higher continuous baseline NLR was independently associated with an increased hazard of BM (HR: 1.77, 95% CI: 1.48–2.12; $p < 0.001$), alongside adenocarcinoma histology (HR: 2.96, 95% CI: 1.41–6.24; $p = 0.004$). In the Stage III sub-cohort sensitivity analysis, continuous baseline NLR maintained its independent prognostic significance (HR: 1.74, 95% CI: 1.44–2.09; $p < 0.001$). High baseline NLR (> 2.26) was significantly associated with a higher cumulative incidence of BM (Gray's test $p < 0.001$), inferior overall survival (median 34.0 vs. 65.0 months; $p = 0.015$), and worse progression-free survival (median 18.0 vs. 42.0 months; $p = 0.006$). Baseline PLR was not significantly associated with BM ($p = 0.136$).

Conclusions

Continuous baseline NLR serves as a readily accessible, independent biomarker for predicting the risk and accelerated onset of brain metastasis in patients with NSCLC undergoing definitive chemoradiotherapy. Baseline NLR may aid in risk-stratified neuro-radiological surveillance protocols.

1. INTRODUCTION

Lung cancer remains the most frequently diagnosed malignancy and the leading cause of cancer-related mortality worldwide, accounting for approximately 2.5 million new cases and 1.8 million deaths annually (1). Non-small cell lung cancer (NSCLC) constitutes approximately 80–85% of all these cases. Given that a substantial proportion of patients present with locally advanced, unresectable disease at the time of diagnosis, definitive concurrent chemoradiotherapy (cCRT)—now routinely followed by consolidation durvalumab—has been established as the standard of care for eligible patients (2). Despite significant advancements in modern radiotherapy (RT) techniques and systemic therapeutics, long-term survival outcomes for patients with locally advanced NSCLC remain suboptimal. In this patient population, the primary driver of treatment failure and mortality is distant metastasis rather than locoregional recurrence (3).

Brain metastasis (BM) represents one of the most frequent and major patterns of distant failure in the clinical course of NSCLC (4). Even when robust locoregional control is achieved via cCRT, a significant fraction of patients ultimately develop BM during follow-up, which significantly decreases both overall survival and quality of life. Consequently, the early identification of patients at high risk for BM prior to treatment initiation is of critical clinical importance. Such predictive stratification could facilitate highly individualized therapeutic strategies, primarily by guiding more rigorous, risk-adapted neuro-radiological surveillance and enabling the early consideration of central nervous system (CNS)-penetrating targeted agents.

In recent years, it has become widely recognized that the interaction between the tumor microenvironment (TME) and the host's systemic immune response plays a pivotal role in driving tumor progression and the metastatic cascade (5). Cancer-associated systemic inflammation facilitates metastasis by promoting angiogenesis, remodeling the extracellular matrix, and fostering an immunosuppressive milieu (4). To quantify this inflammatory response, the neutrophil-to-lymphocyte ratio (NLR) and platelet-to-lymphocyte ratio (PLR)—readily derived from routine peripheral blood counts—have emerged as cost-effective, non-invasive, and robust prognostic biomarkers. Pathophysiologically, neutrophils and platelets are known to secrete pro-angiogenic factors and shield circulating tumor cells from immune surveillance, thereby enhancing their survival and extravasation (6–7). Conversely, lymphocytes coordinate the core anti-tumor cytotoxic immune response. Accordingly, elevated NLR and PLR indicate a shift toward an immunosuppressive and pro-tumorigenic microenvironment, which has been consistently correlated with an increased risk of metastasis and diminished overall survival (7).

While the general prognostic significance of NLR in NSCLC is well-documented, its specific utility in predicting the rate, cumulative incidence, and temporal dynamics of brain metastasis—particularly within

a homogeneously monitored cohort of patients undergoing definitive thoracic radiotherapy—remains inadequately defined. Therefore, this study aimed to evaluate the prognostic value of baseline NLR for brain metastasis occurrence and timing, incorporating time-dependent discrimination metrics and a Stage III surveillance-homogeneous sensitivity analysis.

2. MATERIALS AND METHODS

2.1. Patient Selection and Cohort Definition

The medical records of 722 patients with a diagnosis of non-small cell lung cancer (NSCLC) who received curative-intent RT utilizing the TomoTherapy® HD platform at the Department of Radiation Oncology, xxxxxxx Faculty of Medicine, between January 1, 2014, and December 31, 2024, were retrospectively reviewed.

Patients were deemed eligible for inclusion in the study if they met the following criteria: age ≥ 18 years; pathologically confirmed diagnosis of NSCLC; absence of clinical or radiological evidence of brain metastasis prior to treatment (confirmed via baseline contrast-enhanced brain MRI for Stage III patients, and clinical/radiological evaluation for Stage I–II disease); clinical Stage I–III disease; and receipt of definitive-intent intensity-modulated radiotherapy (IMRT) to a total dose of 60–66 Gy in 30–33 fractions, delivered with either concurrent or sequential chemotherapy. All patients were treated with intensity-modulated radiotherapy (IMRT) or helical TomoTherapy® adhering to standard institutional thoracic dose-volume constraints, ensuring homogenous target coverage and normal tissue sparing across the cohort. The inclusion of early-stage (Stage I-II) patients receiving conventionally fractionated RT (60–66 Gy) rather than Stereotactic Body Radiotherapy (SBRT) reflects the institutional treatment protocols prevalent during the earlier years of this extended study period, prior to the widespread accessibility and adoption of SBRT as the standard of care for medically inoperable early-stage disease at our center. Similarly, because the vast majority of the cohort was treated in the pre-immunotherapy era, the number of patients receiving consolidation durvalumab was extremely limited; thus, this variable was excluded from the primary statistical models to maintain cohort homogeneity. Furthermore, it was strictly required that patients possessed baseline, treatment-naïve complete blood count (CBC) data—specifically absolute neutrophil, lymphocyte, and platelet counts—obtained within a maximum of two weeks prior to the initiation of any oncological intervention (i.e., induction chemotherapy or chemoradiotherapy). To address potential variations in baseline systemic inflammation introduced by induction chemotherapy, the receipt of induction chemotherapy was explicitly incorporated as a covariate in all multivariable survival models, and baseline NLR levels were compared between patients receiving and not receiving induction therapy to assess whether baseline systemic inflammation differed according to treatment sequence. To ensure consistency across the study period, all patients were retrospectively re-staged according to the 9th edition of the International Association for the Study of Lung Cancer (IASLC) TNM staging system based on their baseline clinical and radiological data.

Patients were excluded from the analysis if they had incomplete medical records, irregular oncological follow-ups, or a history of a secondary malignancy. Crucially, to prevent the confounding of systemic inflammatory markers, patients presenting with an active infection, a concurrent chronic inflammatory disease, or a history of immunosuppressive drug use at the time of baseline blood sampling were also excluded. Ultimately, a total of 138 patients who fulfilled all predefined criteria were included in the final study cohort. The detailed step-by-step patient selection process, including the specific reasons for exclusion, is illustrated in the study flowchart (Fig. 1).

2.2. Endpoints and Surveillance Protocol

The primary endpoint of the study was the development of brain metastasis following definitive treatment. In accordance with our institutional clinical protocol, routine screening for asymptomatic brain metastases was not routinely performed in early-stage (Stage I–II) disease. However, for patients with locally advanced Stage III disease, brain surveillance was systematically performed using contrast-enhanced brain magnetic resonance imaging (MRI) both prior to treatment initiation and every three months during post-treatment follow-up, regardless of the presence of neurological symptoms. For early-stage (Stage I–II) disease, post-treatment neuroimaging was conducted upon clinical evaluation or the emergence of neurological symptoms. Consequently, the date of brain metastasis was strictly defined as the date of radiological confirmation (either via routine surveillance or symptom-driven neuroimaging). Secondary endpoints included the association of pre-treatment NLR value with overall survival (OS), progression-free survival (PFS), the number of brain metastatic foci, and the time to brain metastasis. For all time-to-event analyses (including OS, PFS, and time to brain metastasis), 'time zero' was strictly defined as the date of initial pathological diagnosis. Accordingly, the hematological indices analyzed represent the true baseline (pre-treatment) systemic inflammatory status prior to any oncological intervention, including induction chemotherapy. Based on clinical and radiological follow-up, the study cohort was stratified into two distinct groups: patients who developed brain metastasis (n = 40) and those who did not (n = 98).

2.3. Statistical Analysis

The normality of continuous data distribution was assessed utilizing the Kolmogorov-Smirnov and Shapiro-Wilk tests. Given the non-parametric distribution of the data, continuous variables were summarized as medians with their corresponding minimum-maximum values or interquartile ranges (IQR). Categorical variables were expressed as frequencies (n) and percentages (%). Comparisons of baseline demographic and clinical characteristics between patients who developed brain metastasis and those who did not were performed using the Mann-Whitney U test for continuous variables. For categorical variables, the Pearson Chi-square test or Fisher’s exact test was applied, as appropriate.

Given the time-to-event nature of brain metastasis, time-dependent Receiver Operating Characteristic (ROC) curve analysis was employed to evaluate the dynamic predictive performance of pre-treatment NLR at 12, 24, and 28 months (corresponding to the median follow-up period). The optimal threshold (cut-off) value for NLR was established based on the Youden Index derived at the median follow-up time.

For PLR, conventional ROC curve analysis was utilized. The discriminative accuracy was reported as the Area Under the Curve (AUC) along with 95% confidence intervals (CI). To assess model internal stability and optimistic bias, internal validation via 1,000 bootstrap resamples was conducted. For survival visualization (Kaplan-Meier estimates), this ROC-derived cut-off value was utilized to stratify cohorts.

Because death without prior brain metastasis was considered a competing event, cumulative incidence functions were estimated and compared using Gray's test. Cause-specific Cox regression was subsequently employed to evaluate independent clinical and biological predictors associated with the instantaneous hazard of brain metastasis. The proportional hazards assumption was visually and statistically verified using Schoenfeld residuals. To minimize model overfitting given the 40 brain metastasis events, a parsimonious multivariable Cox regression model was constructed across four key clinical and biological covariates (Continuous pre-treatment NLR remained independently associated with the cause-specific hazard of brain metastasis). Accounting for two degrees of freedom for histological subtype, the model estimated five total parameters, maintaining an events-per-variable (EPV) ratio of 8.0 (40 events / 5 degrees of freedom), which satisfies established standards (EPV ≥ 5–10) for survival model stability. Because only 30 patients had stage I–II disease, stage was analyzed as a binary variable (stage I–II vs. III) to reduce the risk of sparse-data bias. To visually evaluate the exposure-response relationship between continuous inflammatory markers and the hazard of brain metastasis, restricted cubic spline (RCS) curves with three knots were generated using the 'rms' package. Because the vast majority of patients had an ECOG performance status of 0 or 1, ECOG PS was omitted from the multivariable Cox regression analysis. Although it showed a statistically significant difference between groups (p = 0.023), this variable was excluded due to severe category imbalance and sparse data (only 3 patients [2.2%] had ECOG PS ≥ 2), as well as to adhere strictly to the events-per-variable (EPV) principle for preventing model overfitting given the number of observed events (n = 40). Results were reported as Hazard Ratios (HR) with corresponding 95% confidence intervals (CI).

Overall survival (OS) and progression-free survival (PFS) were estimated using the Kaplan-Meier method and compared between cohorts using the log-rank test. Conversely, for the brain metastasis endpoint—where death without intracranial progression represents a competing risk—cumulative incidence was calculated using Gray's test, and independent predictors were evaluated via cause-specific Cox proportional hazards regression. For OS analysis, patients who remained alive at the time of the last clinical contact were censored. For PFS, the event was defined as disease progression at any site or death from any cause, and patients without these events were censored at the date of their last radiological or clinical assessment. The cumulative incidence of brain metastasis was estimated by accounting for death as a competing risk, and differences between the inflammatory marker cohorts were compared using Gray's test.

For patients who developed brain metastasis, the number of metastatic foci was categorized as an ordinal variable (1, 2–3, 4–9, and ≥ 10 foci). The association between these categories and baseline NLR levels was analyzed using the Kruskal-Wallis test. Standard statistical analyses were performed using IBM SPSS Statistics version 27.0 (IBM Corp., Armonk, NY, USA). Advanced survival analyses, including

competing risk assessments and restricted cubic spline modeling, were conducted using EZR (Easy R) software, a graphical user interface for R (The R Foundation for Statistical Computing, Vienna, Austria). A two-sided *p*-value of < 0.05 was considered to indicate statistical significance.

2.4. Sensitivity Analysis

To address potential residual confounding related to anatomical disease burden, evaluate lead-time bias, and account for unmeasured baseline asymptomatic brain metastases in early-stage patients who did not undergo routine baseline brain MRI, a pre-specified sensitivity analysis was restricted exclusively to the homogeneous Stage III sub-cohort (n = 108), all of whom underwent mandatory baseline contrast-enhanced cranial MRI. The primary multivariable cause-specific Cox model was re-estimated within this sub-cohort to confirm whether the independent prognostic association of continuous baseline NLR persisted under stringent anatomical homogeneity.

3. RESULTS

3.1. Baseline Clinical and Demographic Characteristics

A total of 138 patients with NSCLC who met the eligibility criteria were included in the final analysis. The mean age of the study cohort was 61.32 ± 8.94 years, with a median age of 62 years (range, 32–79). The patient population was predominantly male (n = 122, 88.4%), and a vast majority had a history of active or former smoking (n = 123, 89.1%). The baseline demographic and clinical characteristics of the patients, stratified by the development of brain metastasis, are summarized in Table 1. The distribution of patients according to the TNM staging system is detailed in Table 2. The median follow-up time for the entire cohort was 28.0 months (interquartile range [IQR]: 16.0–50.2).

Table 1
Clinical and demographic characteristics of the patient groups.

Variable	Brain Metastasis (n = 40)	No Brain Metastasis (n = 98)	*p*-value
Age (years), median (range)	64 (36–78)	60 (32–79)	0.111
NLR, median (range)	3.21 (1.63–11.55)	2.22 (0.75–4.39)	**< 0.001**
PLR, median (range)	150.7 (80.7–600.0)	130.2 (58.6–447.0)	0.136
Gender, n (%)			
Male	35 (87.5%)	87 (88.8%)	0.778
Female	5 (12.5%)	11 (11.2%)	
ECOG PS, n (%)			
0–1	37 (92.5%)	98 (100%)	**0.023**
≥ 2	3 (7.5%)	0 (0%)	
Histological Subtype, n (%)			
SCC	13 (32.5%)	61 (62.2%)	**0.005**
Adenocarcinoma	20 (50.0%)	25 (25.5%)	
Other	7 (17.5%)	12 (12.2%)	
Smoking History, n (%)			
Yes	33 (82.5%)	90 (91.7%)	0.220
No	7 (17.5%)	8 (8.3%)	
T Stage, n (%)			
T1	5 (12.5%)	8 (8.2%)	0.430
T2	7 (17.5%)	23 (23.5%)	
T3	14 (35.0%)	24 (24.5%)	
T4	14 (35.0%)	43 (43.8%)	
N Stage, n (%)			
N0	7 (17.5%)	20 (20.4%)	0.599

Abbreviations: CAD, coronary artery disease; **COPD**, chronic obstructive pulmonary disease; **CT**, computed tomography; **ECOG PS**, Eastern Cooperative Oncology Group performance status; **NLR**, neutrophil-to-lymphocyte ratio; **PET-CT**, positron emission tomography-computed tomography; **PLR**, platelet-to-lymphocyte ratio; **SCC**, squamous cell carcinoma. *Bold p-values indicate statistical significance.*

Variable	Brain Metastasis (n = 40)	No Brain Metastasis (n = 98)	*p*-value
N1	2 (5.0%)	12 (12.2%)	
N2	20 (50.0%)	44 (44.9%)	
N3	11 (27.5%)	22 (22.4%)	
Staging Modality, n (%)			
PET-CT	39 (97.5%)	96 (98.0%)	1.000
CT	1 (2.5%)	2 (2.0%)	
Induction Chemotherapy, n (%)			
Yes	19 (47.5%)	58 (59.2%)	0.265
No	21 (52.5%)	40 (40.8%)	
Diabetes Mellitus, n (%)			
Yes	10 (25.0%)	25 (25.5%)	0.950
No	30 (75.0%)	73 (74.5%)	
Hypertension, n (%)			
Yes	8 (20.0%)	20 (20.4%)	0.957
No	32 (80.0%)	78 (79.6%)	
CAD, n (%)			
Yes	9 (22.5%)	18 (18.4%)	0.579
No	31 (77.5%)	80 (81.6%)	
COPD, n (%)			
Yes	4 (10.0%)	11 (11.2%)	1.000
No	36 (90.0%)	87 (88.8%)	

Abbreviations: CAD, coronary artery disease; **COPD**, chronic obstructive pulmonary disease; **CT**, computed tomography; **ECOG PS**, Eastern Cooperative Oncology Group performance status; **NLR**, neutrophil-to-lymphocyte ratio; **PET-CT**, positron emission tomography-computed tomography; **PLR**, platelet-to-lymphocyte ratio; **SCC**, squamous cell carcinoma. *Bold p-values indicate statistical significance.*

Table 2
Initial clinical stages of the patients according to the 9th edition of the IASLC staging system.

Clinical Stage	Number of Patients (n)	Percentage (%)
Stage I	8	5.8
Stage II	22	15.9
Stage III	108	78.3
Total	138	100.0

Over the course of the follow-up period, brain metastasis (BM) developed in 29.0% of the patients (n = 40), whereas the remaining 71.0% (n = 98) did not develop intracranial progression. Histological subtype analysis revealed that within the BM-positive group, the incidence of adenocarcinoma was significantly higher compared to the BM-negative group (50.0%, n = 20 vs.25.5%, n = 25, respectively; $p = 0.005$), whereas squamous cell carcinoma (SCC) was the predominant histology in patients who did not develop BM (62.2%, n = 61 vs.32.5%, n = 13; $p = 0.005$). Overall, 55.7% of patients (n = 77) received induction chemotherapy prior to definitive RT, with no significant difference between the BM-positive and BM-negative cohorts ($p = 0.265$). Because the vast majority of the study cohort was treated prior to the clinical implementation and routine reimbursement of consolidation immunotherapy in our region, only a negligible subset of patients received consolidation durvalumab (n = 9, 6.5%), precluding its inclusion as an independent variable in multivariable survival analysis. An evaluation of baseline systemic comorbidities demonstrated that the prevalence of Diabetes Mellitus (DM), Hypertension (HT), Coronary Artery Disease (CAD), and Chronic Obstructive Pulmonary Disease (COPD) were homogeneously distributed between the two groups (all $p > 0.05$).

3.2. Comparative Analysis of Systemic Inflammatory Markers

Baseline, treatment-naïve NLR and PLR values were evaluated and compared between the study groups. The median NLR in patients who developed BM was 3.21 (range, 1.63–11.55; IQR: 2.43–4.48), whereas it was calculated as 2.22 (range, 0.75–4.39; IQR: 1.86–2.78) in the BM-negative group. This inter-cohort difference was highly statistically significant ($p < 0.001$) **(**Fig. 2**)**. Although baseline NLR significantly differed between patients who received induction chemotherapy and those who did not (median 2.25 vs. 2.62, $p = 0.001$), multivariable analysis demonstrated that its predictive capacity for brain metastasis remains independent of the initial treatment strategy

Regarding PLR levels, the median value was 150.7 (range, 80.7–600.0; IQR: 103.4–195.7) in the BM-positive group compared to 130.2 (range, 58.6–447.0; IQR: 99.9–170.6) in the BM-negative group. Although the PLR was numerically higher in patients who developed BM, this difference did not reach statistical significance ($p = 0.136$).

3.3. Predictive Performance and ROC Curve Analysis

To evaluate the dynamic predictive performance of baseline NLR for the development of brain metastasis, time-dependent Receiver Operating Characteristic (ROC) curve analysis was conducted at 12, 24, and 28 months (median follow-up) (Table 3). The area under the curve (AUC) values for NLR at 12, 24, and 28 months were 0.78, 0.79, and 0.79, respectively, demonstrating a consistent discriminative capacity over time. Utilizing the Youden Index at the median follow-up of 28 months, the optimal time-dependent cut-off value for NLR was identified as 2.26, yielding a high sensitivity of 91.7% and a specificity of 54.6% for predicting brain metastasis (Fig. 3).

Table 3
Time-dependent Receiver Operating Characteristic (ROC) curve analysis of pre-treatment NLR for predicting brain metastasis over time.

Time Point	AUC	Cut-off Value	Sensitivity (%)	Specificity (%)
12 Months	0.78	2.40	94.6	56.2
24 Months	0.79	2.26	91.7	54.6
28 Months (Median)	0.79	2.26	91.7	54.6
Abbreviations: AUC, area under the curve				

Conversely, ROC curve analysis for PLR demonstrated a non-significant predictive performance with an AUC of 0.58 (95% CI: 0.47–0.69, $p = 0.136$). Because PLR failed to achieve statistical significance, no optimal cut-off value was established for this parameter, and subsequent survival models focused strictly on baseline NLR. Internal validation via 1,000 bootstrap resamples demonstrated an optimism-corrected AUC of 0.76, confirming robust internal consistency.

3.4. Identification of Independent Risk Factors for Brain Metastasis

A parsimonious multivariable cause-specific Cox regression model was constructed using five total parameters across four target covariates (continuous NLR, binary clinical stage, binary induction chemotherapy, and 3-category histological subtype [2 degrees of freedom]), yielding an events-per-variable (EPV) ratio of 8.0 (40 events / 5 degrees of freedom). This strictly adheres to recommended parsimony standards (EPV ≥ 5–10) to minimize model overfitting and optimistic bias. Higher continuous pretreatment NLR was independently associated with an increased cause-specific hazard of brain metastasis (HR: 1.77; 95% CI: 1.48–2.12; $p < 0.001$). Specifically, each one-unit increase in baseline NLR was associated with a 77.3% increase in the hazard of developing cranial relapse. Furthermore, histological subtype was identified as an independent risk factor; patients with adenocarcinoma exhibited a 2.964-fold increased risk of brain metastasis compared to the reference squamous cell carcinoma group (HR: 2.964; 95% CI: 1.408–6.239; $p = 0.004$). Neither overall clinical stage ($p = 0.839$) nor the administration of induction chemotherapy ($p = 0.674$) reached statistical significance in the multivariable model (Table 4).

Table 4
Multivariable cause-specific Cox regression analysis for independent predictors of brain metastasis incorporating the competing risk of death.

Variable	HR	95% CI	*p*-value
Pre-treatment NLR (Continuous)	1.77	1.48–2.12	**< 0.001**
Histological Subtype			**0.016**
Squamous Cell Carcinoma	1.00	-	Reference
Adenocarcinoma	2.96	1.41–6.24	**0.004**
Other	1.66	0.61–4.54	0.324
Clinical Stage (Stage III vs. Stage I-II)	0.92	0.41–2.07	0.839
Induction Chemotherapy (Yes vs. No)	1.17	0.57–2.38	0.674

Abbreviations: CI, confidence interval; **NLR**, neutrophil-to-lymphocyte ratio; **HR**, hazard ratio.

Targetable genetic alterations (e.g., EGFR, ALK, ROS1, BRAF) and PD-L1 expression levels were excluded from the multivariate model due to routine clinical testing limitations and an insufficient number of positive cases, which would have compromised the statistical power of the analysis.

To further visualize this continuous risk dynamic, Restricted Cubic Spline (RCS) curves with three knots were generated. The spline models suggested a potential non-linear association, demonstrating a rapid escalation in the hazard for brain metastasis as baseline continuous NLR increased beyond the exploratory threshold (Fig. 4).

3.5. Sensitivity Analysis in Stage III Sub-Cohort

To address potential detection bias introduced by differential baseline and post-treatment cranial MRI surveillance protocols between early and advanced stages, a pre-specified sensitivity analysis was restricted exclusively to patients with Stage III disease (n = 108), all of whom underwent mandatory baseline and routine follow-up contrast-enhanced brain MRI. Within this anatomically homogeneous sub-cohort, continuous baseline NLR maintained its significant independent association with an increased cause-specific hazard of brain metastasis (HR: 1.74; 95% CI: 1.44–2.09; $p < 0.001$). This confirms that the observed prognostic relationship is exceptionally robust and independent of baseline cranial staging protocols or anatomical disease burden.

3.6. Survival and Progression Analyses

Overall survival (OS) and progression-free survival (PFS), designated as the secondary endpoints of the study, were evaluated using the Kaplan-Meier method. Stratification based on the optimal time-dependent NLR cut-off value of 2.26 demonstrated that median OS was 65.0 months (95% CI: 43.0–NA)

in the low NLR cohort compared to 34.0 months (95% CI: 22.0–53.0) in the high NLR cohort (Log-rank p = 0.015) (Fig. 5).

Regarding PFS, the median duration was significantly longer in the low NLR group at 42.0 months (95% CI: 28.0–NA) compared to 18.0 months (95% CI: 15.0–22.0) in the high NLR group. This difference in PFS between the cohorts was highly statistically significant (Log-rank $p = 0.006$).

3.7. Temporal Dynamics of Brain Metastasis Development

Subgroup analyses were conducted to evaluate the temporal dynamics of brain metastasis development following cCRT while accounting for the competing risk of death. Stratification by the optimal baseline NLR threshold (2.26) demonstrated that patients with an elevated pre-treatment NLR exhibited a significantly higher cumulative incidence and significantly accelerated onset of brain metastasis over time compared to those with a low NLR (Gray's test $p < 0.001$; Fig. 6).

3.8. Association Between Baseline Inflammatory Markers and Intracranial Tumor Burden

To investigate the potential correlation between baseline systemic inflammation and metastatic tumor burden, patients who developed brain metastasis were stratified into four distinct categories based on the number of intracranial foci (1, 2–3, 4–9, and ≥ 10 foci). A Kruskal-Wallis analysis revealed no statistically significant difference in pre-treatment NLR values among the distinct categories of intracranial metastatic foci (Kruskal-Wallis H = 1.29, df = 3, $p = 0.732$).

4. DISCUSSION

Rather than merely confirming the established prognostic role of systemic inflammation in lung cancer, the principal strength and novelty of our study lie in demonstrating the specific association between baseline NLR and the cumulative incidence of brain metastasis over time in patients undergoing definitive thoracic radiotherapy. Furthermore, by employing time-dependent ROC analysis and restricting our secondary validation to a Stage III cohort with standardized imaging surveillance, we provide granular temporal metrics regarding when baseline inflammatory status exerts its maximal predictive efficacy. The critical role of systemic inflammation in neoplastic development, tumor progression, and the metastatic cascade has been well delineated in contemporary oncology literature (8–10). While the inflammatory response within the TME suppresses the anti-tumoral efficacy of the immune system, it simultaneously stimulates local angiogenesis, thereby facilitating the intravasation and dissemination of malignant cells to distant organs (11, 12). In this process, neutrophils promote tumor invasion via the secretion of pro-inflammatory cytokines, proteolytic enzymes, and growth factors, whereas lymphocytes primarily attempt to restrict tumor growth through cytotoxic cellular responses (13, 14). The Neutrophil-to-Lymphocyte Ratio (NLR), which reflects the relative proportions of these two major cell populations in peripheral blood, may have potential as a clinically accessible risk-stratification biomarker, indicating a

shift in the balance between pro-tumoral inflammation and anti-tumoral immunity in favor of the tumor (15, 16).

A primary finding of the present study is the demonstration that an elevated pre-treatment NLR acts as an independent risk factor for the development of brain metastasis in patients with NSCLC undergoing definitive chemoradiotherapy. Our multivariable cause-specific Cox regression model demonstrated that higher continuous pretreatment NLR was independently associated with an increased cause-specific hazard of brain metastasis. Specifically, every one-unit increase in baseline NLR significantly elevates the hazard of cranial relapse by 1.77-fold (HR: 1.77; 95% CI: 1.48–2.12; $p < 0.001$), independent of all other clinical and demographic parameters. Furthermore, survival analyses utilizing this optimal exploratory time-dependent threshold (2.26) demonstrated a significant separation in time to brain metastasis onset, effectively identifying a high-risk subgroup within our cohort. These results align with previous investigations by Marschollek et al. (17) and Hu et al. (15), which similarly reported that an elevated NLR significantly amplifies the intracranial metastatic potential in NSCLC cohorts. Although the cut-off value determined in our cohort (2.26) is slightly lower than the widely varying ranges reported in the literature (2.5–5.0), it achieved high sensitivity (91.7%) and robust dynamic diagnostic discrimination (time-dependent AUC: 0.79 at median follow-up) for predicting brain metastasis (15, 17–18). These findings suggest that pretreatment NLR may provide clinically relevant information for identifying patients at increased risk of intracranial relapse.

Our multivariable cause-specific Cox analysis revealed that the predictive value of NLR for brain metastasis remains significant even after adjusting for key clinical covariates, including histological subtype, overall clinical stage, and the receipt of induction chemotherapy. The higher hazard observed among patients with adenocarcinoma is consistent with previously described histology-associated differences in the propensity for intracranial dissemination. A fundamental conceptual question is whether baseline NLR serves as an intracranial site-specific risk factor or merely reflects a more aggressive systemic disease biology associated with inferior overall and progression-free survival. While systemic inflammation undeniably correlates with broader tumor aggressiveness and poor overall prognosis, our multivariable cause-specific Cox model—which appropriately accounted for the competing risk of mortality—demonstrated a persistent, independent association between continuous NLR and the instantaneous hazard of brain metastasis. While our clinical findings do not directly assess molecular pathways, one possible biological explanation, supported by preclinical and translational literature, is that systemic inflammation may foster a pro-metastatic vascular and immunosuppressive microenvironment that facilitates circulating tumor cell extravasation across the blood-brain barrier.

Crucially, the robustness of our findings is further reinforced by the pre-specified sensitivity analysis restricted exclusively to Stage III patients (n = 108). Because early-stage patients did not undergo routine baseline brain MRI or scheduled post-treatment neuroimaging, differential surveillance intensity could theoretically introduce detection bias or lead-time artifacts. However, continuous baseline NLR maintained a consistent and independent association with the hazard of intracranial relapse within this fully brain-surveilled sub-cohort (HR: 1.74; 95% CI: 1.44–2.09; $p < 0.001$). These findings support the

robustness of the observed association and suggest that differential neuroimaging surveillance alone is unlikely to fully explain the association.

Importantly, the predictive value of baseline NLR remained robust after adjusting for the receipt of induction chemotherapy ($p = 0.674$), confirming that the observed association was not confounded by prior systemic cytoreductive treatments. Biologically, this suggests that even an anatomically lower-stage tumor can harbor a high risk of cranial metastasis if it successfully disrupts the host's systemic immuno-inflammatory profile. Importantly, by strictly constraining our multivariable model to an 8:1 events-per-variable ratio (5 total estimated parameters for 40 events), was intended to limit model complexity and reduce the risk of overfitting, further establishing that the observed predictive power of continuous baseline NLR is biologically genuine rather than a statistical artifact. From a clinical perspective, it is important to contextualize these findings within the evolving therapeutic landscape of Stage III NSCLC. Because our study population reflects a predominantly pre-immunotherapy cohort—with only 6.5% (n = 9) receiving consolidation durvalumab—the observed risk dynamics specifically pertain to patients undergoing definitive chemoradiotherapy alone. Given that immune checkpoint inhibition significantly alters systemic immune homeostasis and intracranial progression patterns, caution should be exercised when extrapolating these results to modern Stage III cohorts treated according to the current durvalumab consolidation standard of care.

Conversely, although elevated PLR has been associated with poor prognosis in various malignancies, our comparative analysis did not demonstrate a statistically significant relationship between baseline PLR and the incidence of brain metastasis ($p = 0.136$). In this cohort, NLR demonstrated a stronger association with clinically detected brain metastasis than PLR, highlighting its potential utility as a more robust circulating marker for risk stratification. (15, 17).

Regarding the prognostic value of systemic inflammatory indices, our data confirmed that an elevated NLR is significantly associated with decreased overall survival (OS) in patients with NSCLC. The median survival time in the high NLR cohort (34.0 months) was significantly shorter compared to those with low baseline markers (65.0 months). These findings corroborate large-scale meta-analyses in the literature linking elevated inflammatory parameters to poor prognosis in NSCLC (19–21). The negative prognostic association of NLR on survival is likely driven by the oncological manifestation of neutrophilia driven by aggressive cytokine release, coupled with lymphopenia secondary to systemic immunosuppression (18, 22–23). Furthermore, elevated baseline NLR was significantly associated with inferior PFS ($p = 0.006$), reinforcing the adverse impact of these inflammatory parameters on disease control and corroborating major studies in the literature (19, 21).

A notable finding of this study regarding cranial metastasis dynamics is that while elevated inflammatory ratios significantly accelerate the time to brain metastasis onset ($p < 0.001$), they do not influence the ultimate number of metastatic foci that develop within the brain. This indicates that while systemic immune dysregulation (high NLR) facilitates and accelerates the extravasation of circulating tumor cells across the blood-brain barrier, the actual number of macroscopic foci established by these cells is

dictated more by the brain microenvironment and the tumor's intrinsic clonal heterogeneity than by baseline systemic parameters (15, 24). To our knowledge, this is one of the few studies specifically focusing on baseline NLR as a predictive marker for brain failure kinetics in definitive thoracic RT settings, rather than broad clinical outcomes like OS or PFS. The implementation of time-dependent AUC metrics allowed us to capture the dynamic predictive value of NLR across different post-treatment time intervals, while the Stage III sub-analysis ensured that our findings were not confounded by heterogeneous follow-up protocols or advanced baseline disease stage.

Several limitations of this study should be acknowledged. First, the retrospective, single-center design and reliance on baseline blood counts may not capture dynamic inflammatory fluctuations. Second, baseline cranial MRI was not routinely mandated for early-stage (Stage I–II) patients—who represented only 21.7% (n = 30) of the overall cohort—whereas all Stage III patients (78.3%, n = 108) underwent mandatory baseline contrast-enhanced MRI. Consequently, the potential presence of pre-existing, asymptomatic baseline micro-metastases in a minor fraction of early-stage patients cannot be entirely excluded, introducing a potential lead-time bias for time-to-event analyses. Crucially, to directly counter this potential lead-time bias, our sensitivity analysis restricted strictly to Stage III patients (n = 108)—all of whom underwent mandatory baseline contrast-enhanced cranial MRI—retained the independent predictive significance of NLR ($p < 0.001$), reinforcing the ultimate validity of our primary conclusions. Third, due to the retrospective timeframe of this study, molecular profiling for targetable driver alterations (e.g., EGFR, ALK, ROS1) was incomplete, precluding their inclusion in multivariable survival models. Consequently, because specific driver alterations in adenocarcinoma are well known to confer enhanced central nervous system tropism, residual confounding by unmeasured molecular subtypes cannot be entirely excluded when evaluating the independent association between baseline NLR and brain metastasis risk. While internal bootstrap validation confirmed acceptable optimism-corrected discrimination (AUC: 0.76), the 2.26 cut-off derived via the Youden Index remains sample-specific and subject to potential selection optimism. Therefore, this threshold should be interpreted as an exploratory benchmark, and external validation in independent prospective cohorts is strictly required before establishing a standardized clinical cut-off for decision-making. Furthermore, while post-treatment cranial imaging was conducted systematically every three months for Stage III patients, routine post-treatment brain surveillance was not performed in early-stage (Stage I–II) patients unless clinically indicated or symptoms developed. Nevertheless, this approach aligns with our primary clinical focus, which is to predict the development of clinically meaningful, symptomatic brain metastases that directly impact patient survival and quality of life. Finally, because locoregional control rates were not explicitly analyzed, we cannot definitively rule out that a proportion of subsequent cranial relapses might be secondary to unmeasured locoregional treatment failures rather than primary isolated systemic seeding. Of note, our findings do not provide direct evidence to justify routine prophylactic interventions; instead, they provide a hypothesis-generating risk stratification for future evaluation of intensified CNS surveillance and risk-adapted neuro-radiological monitoring.

5. CONCLUSION

Derived from routine pre-treatment peripheral blood counts, continuous baseline Neutrophil-to-Lymphocyte Ratio (NLR) acts as an independent risk-associated biomarker for the risk and accelerated onset of brain metastasis in patients with NSCLC undergoing definitive chemoradiotherapy. Rather than advocating for a fixed clinical cut-off or immediate prophylactic cranial interventions, our findings support the exploratory use of baseline NLR as a cost-effective stratification tool to identify high-risk patients who may benefit from intensified neuro-radiological surveillance. Prospective, multi-institutional studies are needed to externally validate these predictive thresholds before integration into standardized clinical pathways.

Declarations

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Data Availability Statement

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

Declaration of Generative AI and AI-assisted technologies, if any

The authors used ChatGPT (OpenAI) solely for English language editing and improvement of readability. All scientific content, data analysis, interpretation of the results, and final manuscript preparation were performed and verified by the authors, who take full responsibility for the content of this manuscript.

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Informed Consent

The requirement for informed consent was waived owing to the retrospective design of the study and the use of anonymized patient data.

Conflicts of Interest

All authors have completed the ICMJE uniform disclosure form. The authors have no conflicts of interest to declare.

Ethics Approval

The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. The study was conducted in

accordance with the Declaration of Helsinki and its subsequent amendments. The study was approved by the Non-Interventional Clinical Research Ethics Committee of Dicle University Faculty of Medicine (Decision No. 155; March 19, 2025) and individual consent for this retrospective analysis was waived.

Author Contribution

S.T. contributed to the conception and design of the study, data analysis and interpretation, and wrote the main manuscript text. S.T., C.T., and Z.U. provided study materials and patients. Administrative support was provided by S.T. and C.T. Data collection and assembly were performed by İ.K., C.T., and O.İ. All authors participated in drafting or revising the manuscript and gave final approval of the version to be published.

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Figures

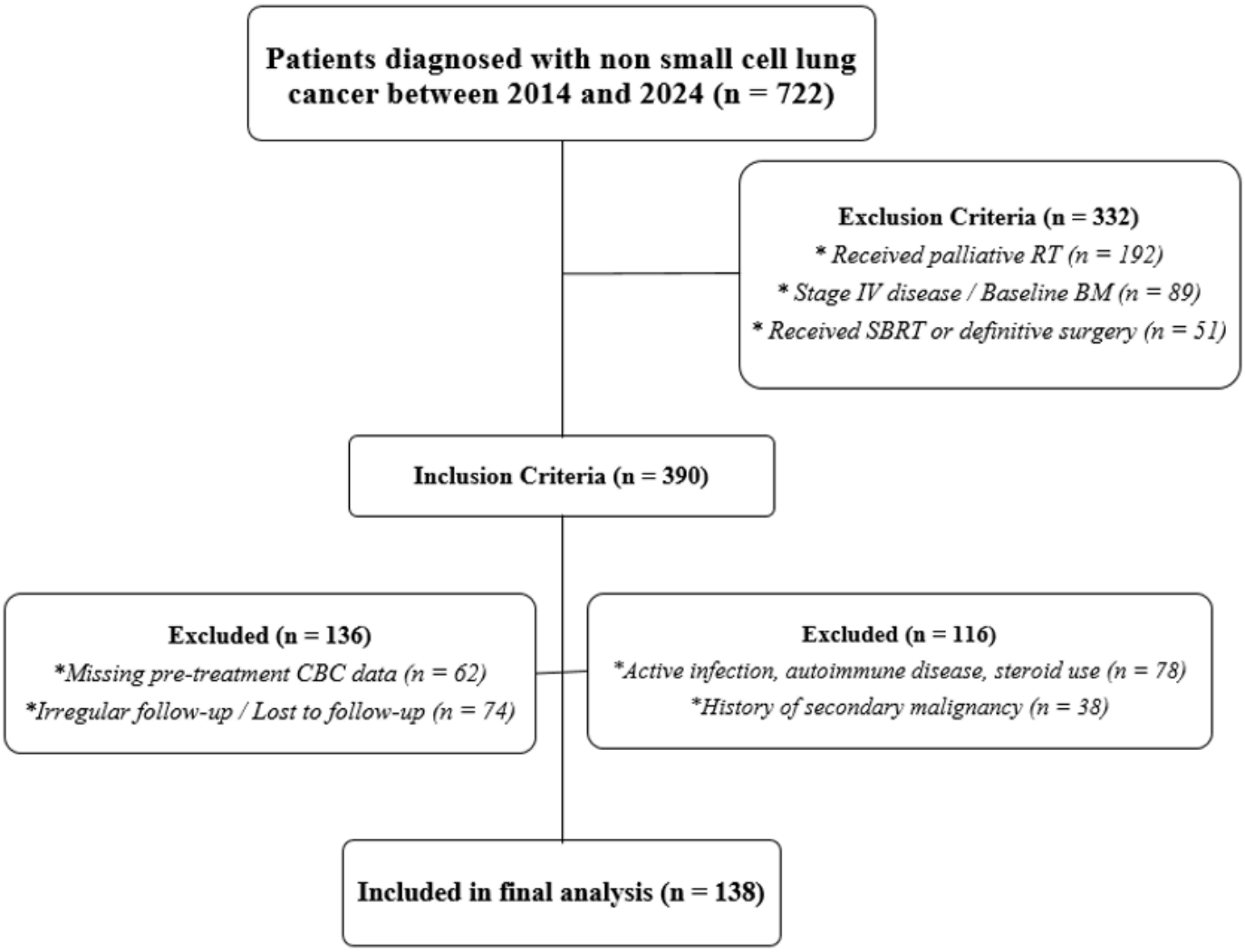


Figure 1

Study flowchart detailing the patient selection process and exclusion criteria.

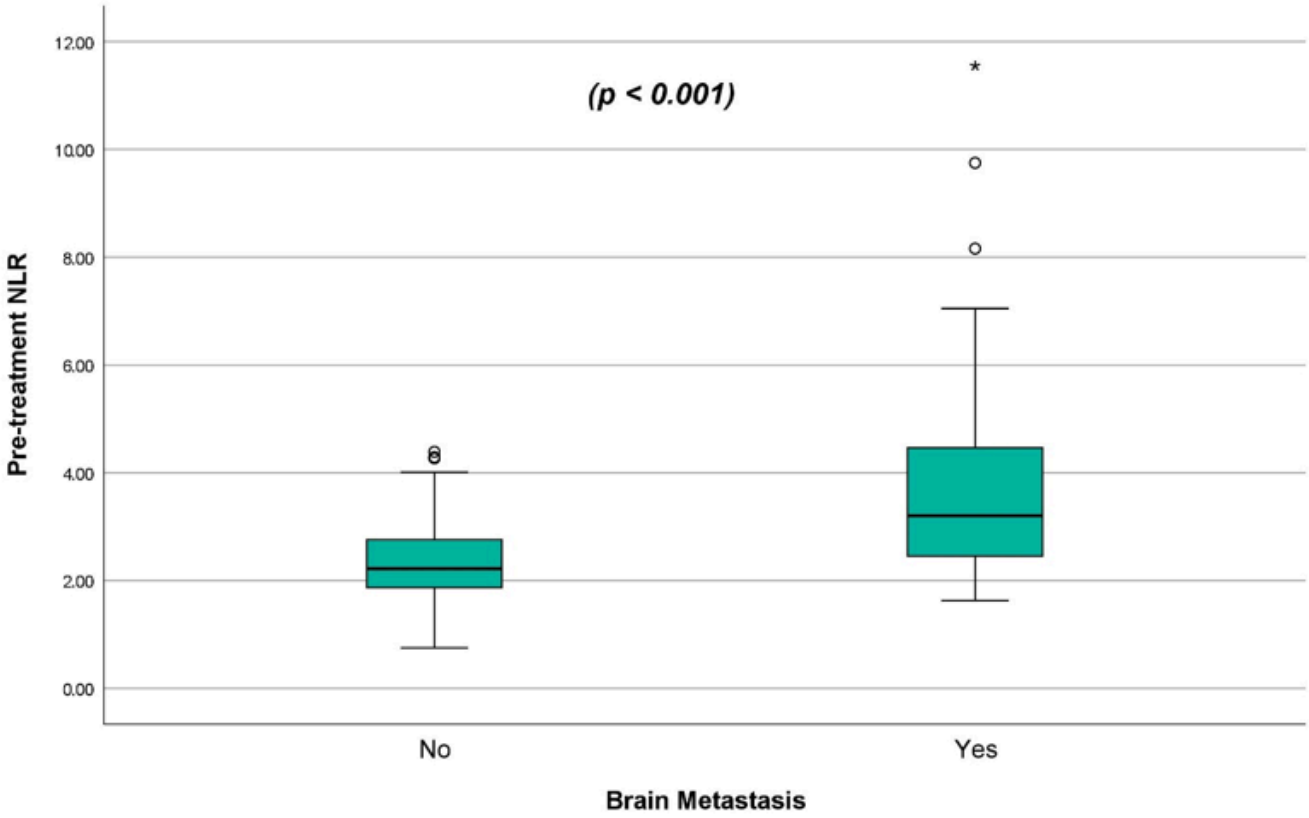


Figure 2

Distribution of pre-treatment NLR by brain metastasis status.

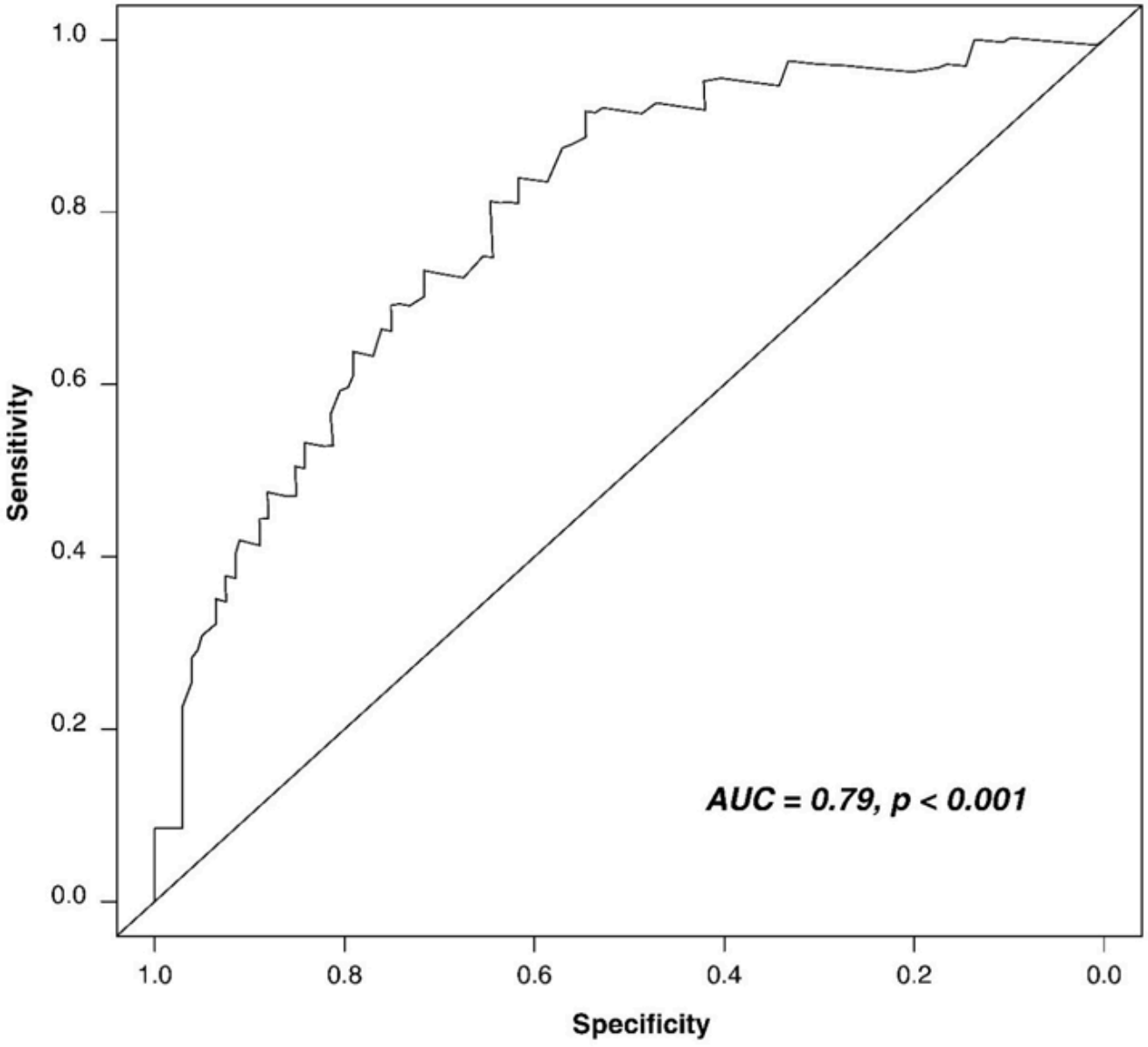


Figure 3

Time-dependent ROC curve at the median follow-up (28 months) demonstrating the dynamic diagnostic value of pre-treatment Neutrophil-to-Lymphocyte Ratio (NLR) in predicting brain metastasis. (*AUC: 0.79, optimal cut-off: 2.26*).

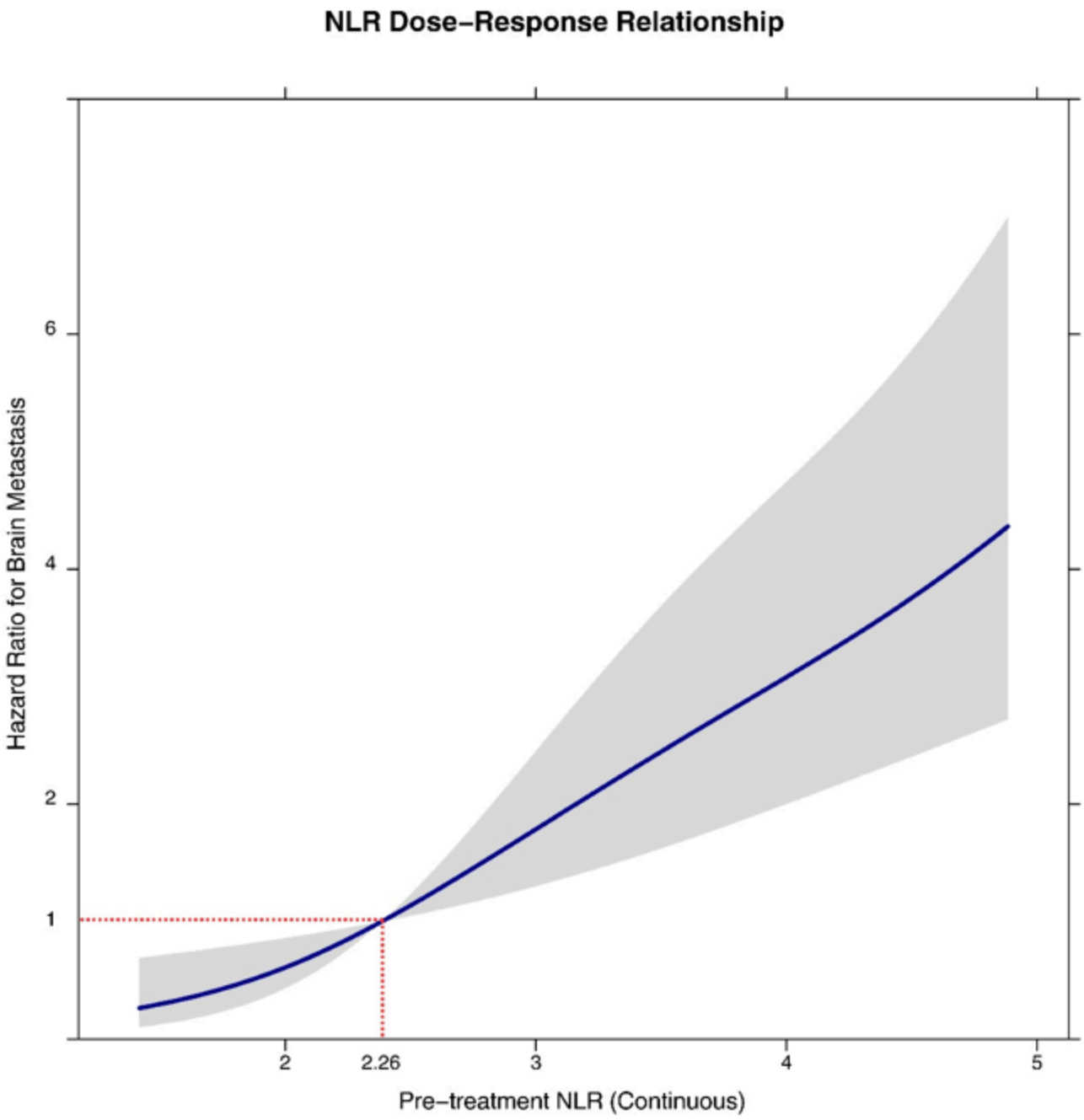


Figure 4

Restricted cubic spline (RCS) curves demonstrating the continuous relationship between baseline pre-treatment NLR and the hazard ratio for brain metastasis. The red reference lines indicate the hazard ratio of 1.0 and the NLR cut-off value of 2.26. The solid curve represents the estimated hazard ratio, and the shaded grey area indicates the 95% confidence interval.

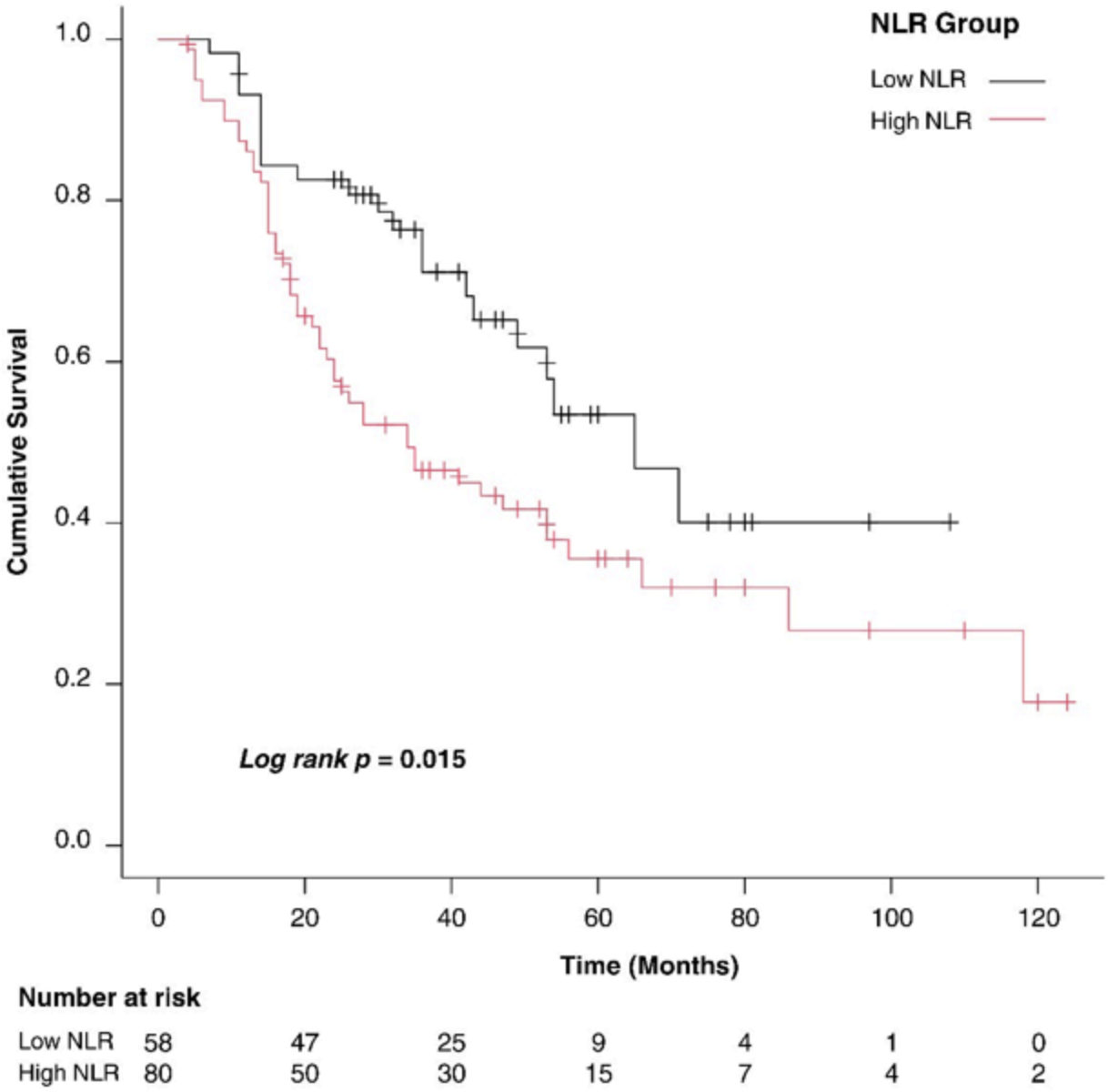


Figure 5

Kaplan-Meier survival curves for overall survival (OS) stratified by pre-treatment NLR (Log-rank p = 0.015). Tick marks represent censored patients.

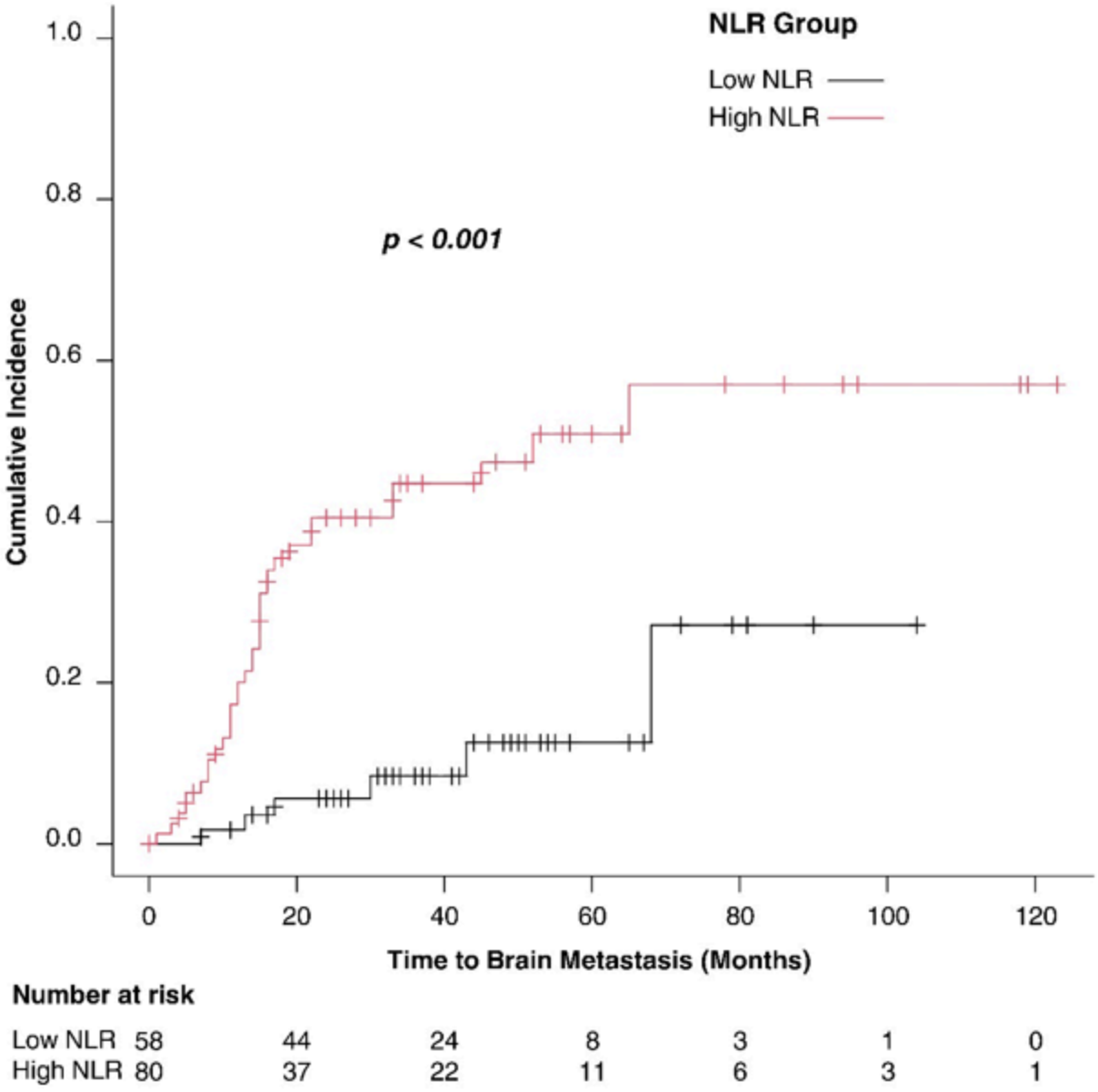


Figure 6

Cumulative incidence curves illustrating the time to brain metastasis, adjusting for the competing risk of death without intracranial progression, stratified by pre-treatment NLR.