Hematologic markers of distant metastases in gastric cancer

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Background: High neutrophil-lymphocyte ratio (NLR) is associated with poor overall survival (OS) in gastric cancer. This study evaluates whether NLR, in addition to other parameters including absolute neutrophil count (ANC), absolute lymphocyte count (ALC), absolute eosinophil count (AEC), absolute monocyte count (AMC), monocyte-lymphocyte ratio (MLR), and platelet-lymphocyte ratio (PLR) are associated with distant metastases, a common and poor prognostic feature of gastric cancer.

Methods: Clinical data from 502 gastric cancer patients treated at King Hussein Cancer Center (Amman, Jordan) have been retrospectively reviewed. We examined the association between ANC, ALC, AEC, AMC, NLR, MLR and PLR with the baseline distant metastases and OS. Receiver operating characteristic (ROC) curve analysis was utilized to determine the optimal NLR cutoff value for association with distant metastases.

Results: Univariate and multivariate analyses showed that patients with high baseline NLR ($\geq$3.9) had more distant metastases on presentation than patients with low NLR (<3.9), (P value: 0.0001 and 0.0005, respectively). Furthermore, patients with high baseline ANC ($\geq$6,015/μL), AEC ($\geq$215/μL), PLR ($\geq$0.15) had more distant metastases in comparison to patients with low baseline ANC (<6,015/μL), AEC (<215/μL), PLR (<0.15) (P value: 0.024, 0.001, and 0.001, respectively). High ANC, NLR, MLR and PLR are associated with poor OS (P value: 0.046, 0.0003, 0.027, and <0.0001, respectively).

Conclusions: High ANC, AEC, NLR, and PLR are associated with distant metastases on presentation in gastric cancer. In the era of cancer immunotherapy, whether these immune phenomena predict the response of gastric cancer to immunotherapy is unknown.

Keywords: Gastric cancer; neutrophil-lymphocyte ratio (NLR); distant metastases

Introduction

Gastric cancer is the second most common gastrointestinal malignancy after colorectal cancer (CRC) with one million cases diagnosed worldwide annually (1). Around 40% of gastric cancer patients have distant metastases at the time of diagnosis (2). Despite the recent advancements in the management of metastatic gastric cancer, the prognosis remains poor with a 5-year survival rate of less than 4% (3).

Inflammation has been linked to carcinogenesis, metastases and survival outcomes in cancer (4,5). However, the direct relationship between inflammation and metastases is unclear. Absolute neutrophil count (ANC), absolute lymphocyte count (ALC), absolute eosinophil count (AEC), absolute monocyte count (AMC), monocyte-lymphocyte ratio (MLR), platelet-lymphocyte ratio (PLR) and neutrophil-lymphocyte ratio (NLR) were found to have a negative impact on survival in various tumors including gastric cancer (6-10). Additionally; recent studies showed that high NLR is an independent predictive factor for baseline presence of brain metastases in advanced non-small cell lung cancer (NSCLC), and for the baseline presence of lung metastases in advanced CRC (11,12).

Various explanations have been proposed to describe the relationship between inflammation and metastases. High neutrophil count resulting in high NLR is thought to promote metastases via releasing certain growth factors such as VEGF and other proteases (13). Alternatively, lymphocytes have an essential role in eliminating tumor cells by inducing cytotoxic cell death and impeding tumor cell proliferation and migration, explaining the association of low ALC with poor outcomes in cancer due to diminished immune response to malignancy (14,15).

In this retrospective study, we aim to further identify the relationship between blood inflammatory cells with distant metastases in gastric cancer as this might have important implications on the management of gastric cancer. Our proposed hypothetical questions were, do gastric cancer patients with high NLR have a high incidence of distant metastases? Do they need more frequent surveillance imaging? Do they benefit from prophylactic strategies? And finally, might they respond better to the immunotherapy?

Methods

This is a retrospective chart review study that was approved by the Institutional Review Board office at King Hussein Cancer Center (IRB Approval No. 18KHCC54). Informed consent is not needed since it is a retrospective chart review study. Five hundred and two patients diagnosed with gastric cancer, who received treatment at KHCC (Amman, Jordan), were included. Positron emission tomography (PET) scans, computed tomography (CT) scans and magnetic resonance imaging (MRI) were used to detect distant metastases. Clinical data including age, gender, location of primary tumor, and the presence of baseline distant metastases were collected and summarized in *Table 1*. Patients were excluded if they were on steroids before obtaining a complete blood count (CBC). Using steroids was expected to confound the results of the study as it leads to leukocytosis (16) and specifically neutrophilia (17).

CBC with differential white cell count at the time of diagnosis was collected before the initiation of any cancer specific treatment (systemic treatment or radiation). The pre-treatment baseline NLR, MLR and PLR were calculated using these formulas: NLR = ANC/ALC, MLR = AMC/ALC and PLR = platelet count/ALC.

The receiver operating characteristic (ROC) curve was used to determine the optimal cut-off value for NLR associated with distant metastases, matching the most extreme joint sensitivity and specificity. The association between NLR and clinical factors including the age, gender, and the location of primary tumor with the presence of distant metastases were examined. Univariate and multivariate logistic regression analyses were used to test the association between the various variables and presence of distant metastases on diagnosis. A P value of ≤0.05 was determined as the cut off value for a significant association.

Our analysis proceeded stepwise. In the first phase, we examined the association between baseline NLR with the presence of distant metastases. In the second phase, we examined the association between other hematologic parameters including ANC, ALC, AEC, AMC, MLR & PLR with the baseline presence of distant metastases. In the third phase, we examined the association between baseline presence of distant metastases with the clinical variables like age, gender, and location of the primary tumor. In the fourth phase, we performed a multivariate analysis that included the collected variables (age, gender, and location of primary tumor) with NLR as a continuous variable, to identify correlation with baseline distant metastases. In the last phase, the fifth, we examined the association between

Table 1 Characteristics of gastric cancer patients

Patients features	No. of patients (%)
Age	
>54	251 (50.0)
≤54	251 (50.0)
Gender	
Male	281 (56.0)
Female	221 (44.0)
Location	
Cardia, fundus & body	143 (29.0)
Antrum & pylorus	75 (15.0)
Lesser and greater curvature	46 (9.0)
Overlapping & unspecified	238 (47.0)
Baseline peritoneum metastases*	
Yes	81 (17.3)
No	391 (82.7)
Baseline liver metastases**	
Yes	69 (14.6)
No	403 (85.4)
Baseline bone metastases***	
Yes	40 (9.0)
No	406 (91.0)
Clinical TNM stage****	
0, I & II	69 (14.0)
III	259 (52.0)
IV	173 (34.0)
Hematologic parameters, mean [median]	
ANC	11,000 [6,015]
ALC	2,800 [2,000]
AEC	360 [215]
AMC	1,000 [660]
NLR	4.4 [3.3]
MLR	0.4 [0.3]
PLR	0.2 [0.15]

*, data on 30 cases was missing; **, data on 30 cases was missing; ***, data on 56 cases was missing; ****, data on 1 case was missing. ANC, absolute neutrophil count; ALC, absolute lymphocyte count; AEC, absolute eosinophil count; AMC, absolute monocyte count; NLR, neutrophil-lymphocyte ratio; MLR, monocyte-lymphocyte ratio; PLR, platelet-lymphocyte ratio.
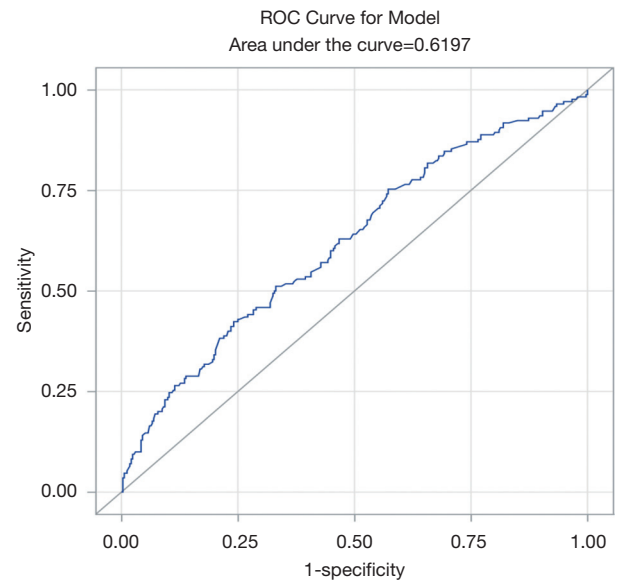

Figure 1 ROC and AUC for NLR. ROC, receiver-operating-characteristic; AUC, area under the curve; NLR, neutrophil-lymphocyte ratio.

the hematologic parameters including ANC, ALC, AEC, AMC, NLR, MLR & PLR with overall survival (OS).

Results

The clinical features of 502 gastric cancer patients are summarized in *Table 1*. More than half of the patients were males (56.0% versus 44.0%, with a male to female ratio of 1.3:1). The median age at diagnosis was 54 years old. The median OS for all patients was 12 months. The baseline NLR mean was 4.4, and the median was 3.3. The optimal NLR cutoff value to correlate with distant metastases was determined to be 3.9 using the ROC curve where the area under curve (AUC) was 0.6197 (*Figure 1*).

The relationship between the various hematologic cell counts, their ratios and the baseline presence of distant metastases are shown in *Table 2*. Patients with high baseline NLR (≥3.9) were more likely to have distant metastases at diagnosis in comparison to patients with low baseline NLR (<3.9) [P=0.0001, odds ratio (OR): 0.5, 95% CI: 0.3–0.7]. High baseline ANC (≥6,015/µL), high AEC (≥215 /µL), and high PLR (≥0.15) were significantly associated with baseline distant metastases (P=0.024, 0.001, and 0.001 respectively). The ROC curve was utilized to determine which parameter including NLR, ANC, AEC, or PLR is a better predictor of baseline presence of distant metastases (*Figure 2*). As shown,

Table 2 The association between hematologic indices with the presence of baseline distant metastasis

Variable	Baseline distant metastases		P value	OR	95% CI
	Present	Absent			
Baseline ANC			0.024	0.65	0.4–0.9
≥6,015	97 (39%)	154 (61%)			
<6,015	73 (29%)	178 (71%)			
Baseline ALC			0.7	1.0	0.7–1.6
≥2,000	83 (33%)	168 (67%)			
<2,000	87 (34%)	164 (66%)			
Baseline AMC			0.19	0.8	0.5–1.1
≥660	92 (37%)	159 (63%)			
<660	78 (31%)	173 (69%)			
Baseline AEC			0.001	0.5	0.3–0.8
≥215	98 (67.6%)	151 (61%)			
<215	47 (25%)	142 (75%)			
Baseline NLR			0.0001	0.5	0.3–0.7
≥3.9	86 (44%)	110 (56%)			
<3.9	84 (27%)	222 (73%)			
Baseline MLR			0.45	0.9	0.6–1.3
≥0.3	89 (35%)	162 (65%)			
<0.3	81 (32%)	170 (68%)			
Baseline PLR			0.001	0.5	0.3–0.8
≥0.15	98 (40%)	150 (60%)			
<0.15	57 (25%)	171 (75%)			

ANC, absolute neutrophil count; ALC, absolute lymphocyte count; AEC, absolute eosinophil count; AMC, absolute monocyte count; NLR, neutrophil-lymphocyte ratio; MLR, monocyte-lymphocyte ratio; PLR, platelet-lymphocyte ratio; OR, odds ratio.

NLR has the highest AUC followed by ANC, PLR, and then AEC which indicates that NLR is a better predictor of distant metastases in comparison to the other tested parameters. Accordingly, NLR was the only parameter that was included in the subsequent multivariate analysis. ALC, AMC, and MLR did not show significant association with the presence of baseline distant metastases in gastric cancer
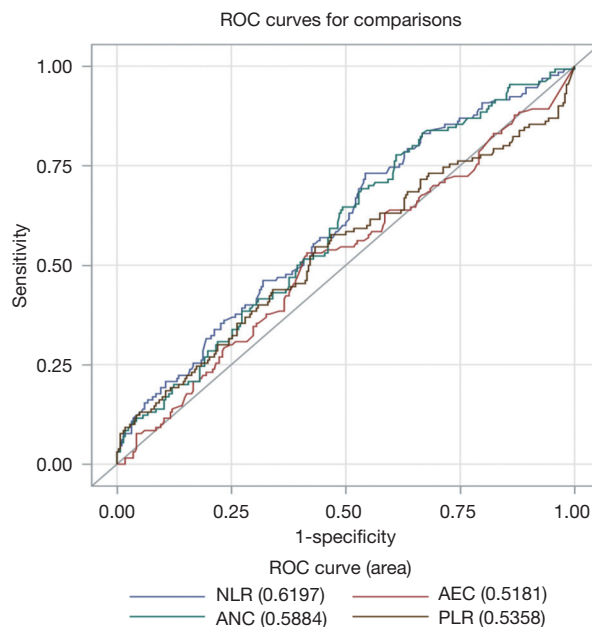

Figure 2 ROC curves of NLR, ANC, PLR, and AEC for predicting distant metastasis. ROC, receiver-operating-characteristic; NLR, neutrophil-lymphocyte ratio; ANC, absolute neutrophil count; PLR, platelet-lymphocyte ratio; AEC, absolute eosinophil count.

(P=0.7, 0.19 and 0.45, respectively).

Table 3 shows the univariate and multivariate analyses assessing the association of NLR and clinical variables with baseline distant metastases. Age was significantly associated with the baseline presence of distant metastases. Younger patients (≤54 years) had more distant metastases compared to older ones (>54 years) (P=0.014). Gender and location of the primary tumor were not associated with baseline presence of distant metastases in the univariate analysis (P=0.32 and 0.33, respectively). Despite of that; gender and location of the primary tumor were included in the multivariate analysis because of their potential clinical relevance. In the multivariate analysis, NLR remained significantly associated with the baseline presence of distant metastases (P=0.0005).

The results of survival analysis are summarized in *Table 4*. High baseline ANC, NLR, MLR, and PLR were associated with poor OS (P=0.0455, 0.0003, 0.0270, and <0.0001, respectively) (*Figures 3-6*). Baseline ALC, AEC, and AMC were not associated with the OS (P=0.07, 0.4, and 0.66,

Table 3 Univariate and multivariate analyses for the association of the different variables with the presence of baseline distant metastases

Variable	Baseline distant metastases		Univariate		Multivariate	
	Present	Absent	P value	OR (95% CI)	P value	OR (95% CI)
Age			0.014		0.021	
>54	72 (29%)	179 (71%)		0.6 (0.4–0.9)		0.6 (0.4–0.9)
≤54	98 (39%)	153 (61%)				
Gender			0.32		0.6	
Female	80 (36%)	141 (64%)		1.2 (0.8–1.7)		1.1 (0.8–1.7)
Male	90 (32%)	191 (68%)				
Location			0.33		0.84	
Cardia, fundus & body	45 (31%)	98 (69%)		0.8 (0.4–1.7)		1.1 (0.35–3.6)
Antrum & pylorus	10 (13%)	65 (87%)				
Lesser and greater curvature	9 (20%)	36 (80%)				
Overlapping & unspecified	109 (46%)	129 (54%)				
Baseline NLR			0.0001		0.0005	
>3.9	86 (44%)	110 (56%)		0.48 (0.31–0.68)		0.5 (0.32–0.74)
≤3.9	84 (27%)	222 (73%)				

NLR, neutrophil-lymphocyte ratio; OR, odds ratio.

Discussion

To our knowledge, this is the first study assessing the potential predictive value of NLR for detecting distant metastases in patients with gastric cancer. An elevated baseline NLR (≥3.9) was an independent factor associated with the baseline presence of distant metastases in gastric cancer as seen in univariate and multivariate analyses after adjusting for important covariates (P=0.0001, and P=0.0005, respectively) (*Table 3*). Additionally; patients with elevated baseline ANC, AEC, and PLR had more distant metastases in comparison to patients with low baseline ANC, AEC, and PLR (P=0.024, 0.001 and 0.001, respectively). Thus, basic hematological parameters might be of value in predicting the baseline presence of distant metastases in patients with gastric cancer.

The presence of distant metastases is the most worrisome prognostic feature of gastric cancer (18). Using simple tests such as CBC to predict the presence of distant metastasis upon diagnosis, and probably predicting the likelihood of subsequent development of distant metastases, might have a direct impact on gastric cancer patients' care. Surveillance for distant metastases through more frequent imaging, and consideration for prophylactic approaches in patients with high NLR could be an attractive area for future research.

Neutrophil to lymphocyte ratio was shown to correlate with PD1/PD-L1 expression, and with response to chemotherapy and immunotherapy in various cancers (19-23). NLR was found to be associated with immune checkpoint molecules PD1\PD-L1 expression in gastric cancer and biliary tract cancer which likely indicates that NLR could be used as a marker for PDL-1 expression in solid tumors (24,25).

Treatment of distant metastases in gastric cancer includes radiation, chemotherapy, immunotherapy, and rarely surgery. Whether NLR is associated with response of the distant metastases to treatment is unknown. A major advancement in the management of gastric cancer will be achieved if NLR is confirmed to be a predictive marker of distant metastases' response to immunotherapy or other available treatments.

Limitations of this study include the retrospective nature of the data with all cases collected from a single cancer center. There was no stratification by the number and sites

534

Abu-Shawer et al. Markers of metastases in gastric cancer

Table 4 The results of survival analysis in relation to the various hematological parameters

Variable	Median of OS (months)	Number of patients	P value	HR	95% CI
ANC			0.046	1.3	1.004–1.6
≥6,015	11	232 (49%)			
<6,015	15	238 (51%)			
ALC			0.07	0.8	0.6–1.005
≥2,000	14	238 (51%)			
<2,000	10	232 (49%)			
AEC			0.4	1.11	0.9–1.4
≥215	12	231 (56%)			
<215	13	180 (44%)			
AMC			0.66	1.055	0.8–1.4
≥660	12	234 (49%)			
<660	13	236 (51%)			
NLR (median)			0.0003	1.5	1.2–2.0
≥3.3	9	223 (47%)			
<3.3	18	247 (53%)			
MLR			0.027	1.3	1.03–1.66
≥0.3	11	235 (50%)			
<0.3	15	235 (50%)			
PLR			<0.0001	1.7	1.3–2.1
≥0.15	10	231 (52%)			
<0.15	22	216 (48%)			

ANC, absolute neutrophil count; ALC, absolute lymphocyte count; AEC, absolute eosinophil count; AMC, absolute monocyte count; NLR, neutrophil-lymphocyte ratio; MLR, monocyte-lymphocyte ratio; PLR, platelet-lymphocyte ratio.

Figure 3 Kaplan Meier curve for overall survival for patients with ANC ≥6,015. ANC, absolute neutrophil count.
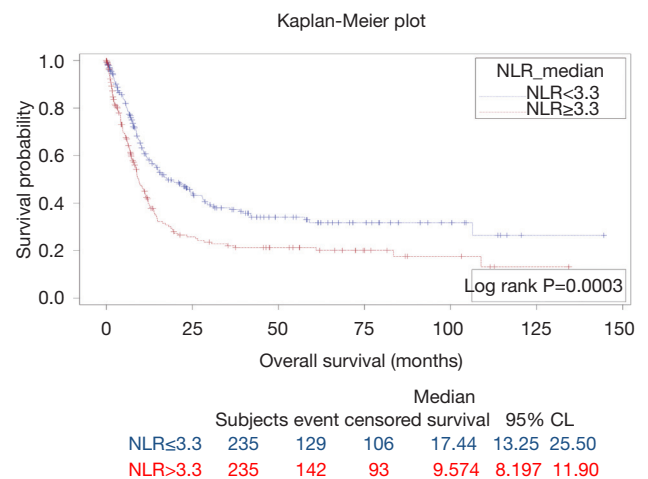
Figure 4 Kaplan Meier curve for overall survival for patients with NLR >3.3. NLR, neutrophil-lymphocyte ratio.

of distant metastatic lesions. There was no assessment of the exact treatments delivered. Additionally, no solid data on the confounding factors that might elevate NLR such as infections were provided.

Conclusions

Elevated baseline NLR is an independent predictive factor for the presence of baseline distant metastases in gastric cancer. This inflammatory-distant metastases association is worthy of further studying in prospective trials, and might be used as a stratification tool to assess response of distant metastases to systemic treatments such as immunotherapy. This might also have implications regarding distant metastases surveillance in patients with high NLR, and probably considering prophylactic approaches in this patient subset.
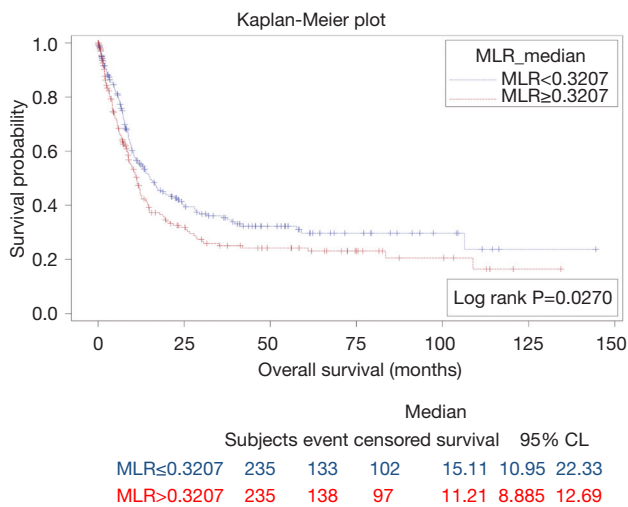
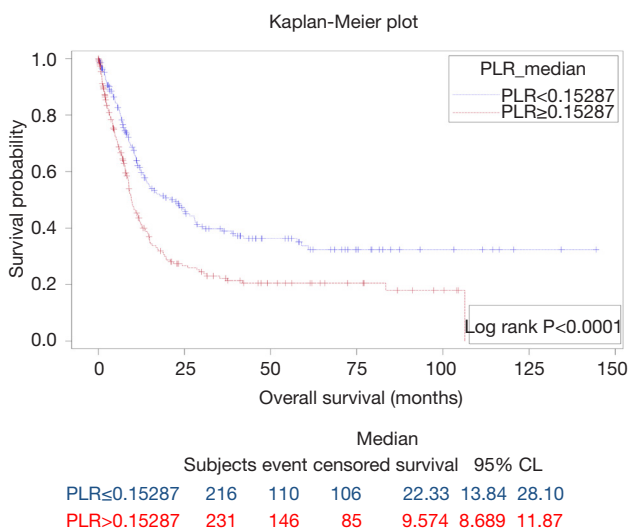**Figure 5** Kaplan Meier curve for overall survival for patients with MLR >0.3. MLR, monocyte-lymphocyte ratio.

Figure 6 Kaplan Meier curve for overall survival for patients with PLR >0.15. PLR, platelet-lymphocyte ratio.

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None.

Footnote

Conflicts of Interest: The authors have no conflicts of interest to declare.

Ethical Statement: The study was approved by the

Institutional Review Board Office at King Hussein Cancer Center (IRB Approval No: 18KHCC54).

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536

Abu-Shawer et al. Markers of metastases in gastric cancer

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