Prognostic Significance of Metastatic Lymph Node Ratio in Patients with Gastric Cancer After Curative Gastrectomy: A Large sample, Single center Retrospective Study

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Abstract

Background:

The aim of this study was to examine the prognostic value of Metastatic lymph node ratio (MLNR) in patients with gastric cancer (GC) after curative gastrectomy and the potential for new indicators to strengthen the current guidelines.

Methods:

We retrospectively examined 3864 patients with GC who underwent curative gastrectomy between February 2011 and February 2016. Patients were classified into low-MLNR (L-MLNR), and high-MLNR (H-MLNR) groups according to adjusted X-tile cutoff-value of 0.25 for MLNR, and their clinicopathological characteristics and survival rates were compared.

Results:

Survival analysis indicated that MLNR was negatively associated with overall survival (OS) ($P < 0.001$) and was an independent prognostic predictor in 3864 GC patients ($P < 0.001$). Stratum analysis and univariate COX proportional hazards analysis were performed to validate the prognostic value of MLNR in various subgroups with clinicopathological characteristics (gender, age at surgery, tumor location, tumor size, type of gastrectomy, T stage, N stage, ELN, perineural invasion, vascular invasion, and TNM stage). MLNR is a prognostic indicator for patients with GC, regardless whether the number of lymph nodes examined is < 16 or ≥ 16 .

Conclusions:

The MLNR may become a new indicator for evaluating prognosis of patients with curative gastrectomy and complement the current guidelines.

Background

Gastric cancer (GC) is the fifth most common malignancy and the third leading cause of cancer-related deaths worldwide. In 2018, there were 103,701 diagnoses and 781,631 disease-related deaths.(1, 2) The Union for International Cancer Control (UICC) TNM staging is the most commonly and extensively prognostic factor for GC, which represents the depth of primary tumor invasion (T), regional lymph node metastases (N), and distant metastases (M) in GC.(3, 4)The TNM staging of the 8th edition is superior to that of the 7th edition in predicting overall survival (OS), but both versions are at risk of staging migration. Staging migration occurs when the number of lymph nodes examined is insufficient. In particular, when it is less than 16, N3b patients may be inappropriately classified as N3a.(5)

It is essential to find a factor independent of TNM classification to better predict the prognosis of GC. In recent years, several studies have indicated that Metastatic lymph node ratio (MLNR) played a significant

role in prognosis in many common cancers, such as breast, colon and rectal cancer.(6, 7) Metastatic Lymph node ratio (MLNR) is defined as the ratio of the number of metastatic lymph nodes to the total number of examined lymph nodes (ELN). (8)Additionally, some investigators have proposed that MLNR could be a simple and useful prognostic predictor in patients with GC.(9, 10)

Therefore, the aim of this study was to examine the prognostic value of Metastatic lymph node ratio (MLNR) in patients with gastric cancer (GC) after curative gastrectomy and the potential for this new indicator to strengthen the current guidelines.

Materials And Methods

Patients and follow-ups

This study retrospectively investigated consecutive patients with GC who underwent gastrectomy. Between February 2011 and February 2016, a total of 3864 patients were included at the First Affiliated Hospital of Anhui Medical University. The primary tumor including GC was staged following the American Joint Committee on Cancer (AJCC) TNM staging system. According to the guidelines of the Japanese Gastric Cancer Association, lymph node dissection was performed for all primary tumors. D1 or D1 + lymphadenectomy for patients with clinical T1 and N0 tumors, and D2 or D2 + lymphadenectomy for patients with clinical T2 or more advanced tumors and those with N1 or more advanced tumors.

The inclusion criteria included:1). Histologically confirmed primary gastric adenocarcinoma in patients;2). Gastrectomy was histologically confirmed as R0;3). Patients had no history of gastrectomy or other malignant tumors;4). Patients had no distant metastases, positive peritoneal cytology, peritoneal dissemination;5). Patients did not have Siewert-I or II tumors at the esophagogastric junction;6). Patients did not receive neoadjuvant radiotherapy;7). Patients survived in the perioperative period;8). Patients had complete medical records and follow-up information;

All 3864 patients were followed up every three or six months for two years after discharge, then every six months for the following three years, and annually thereafter until death. The median follow-up time was 62.7 (range: 1.4 – 110.2) months. The follow-up was completed for all patients in February 2021. This study was approved by the local ethics committee of our hospital. Informed consent was obtained from all patients included in the study.

Selection of cut-off value

The X-tile software (version 3.6.1, Yale University, New Haven, CT) uses the "minimum p-value" method to estimate the optimal cut-off value. The X-tile software was developed by Robert et al. in 2004. X-tile plots can divide a population into two levels (low and high level) for p value estimation. The software provides the relevant Kaplan-Meier curves and displays an "on-the-fly" histogram to the user, with the best p-value obtained by rigorous statistical evaluation of the X-tile.(11, 12)

2.3 Statistical analysis

To assess the significant association between clinicopathological characteristics and patients' survival, univariate and multivariate analyses were performed using the Kaplan-Meier method and Cox proportional hazards analysis. The log-rank test was used to examine the statistical significance of differences among different survival curves. Clinicopathological characteristics significantly related to MLNR were evaluated by the Chi-square test and multinomial logistic regression. The discriminative ability was measured by calculating the Bayesian Information Criterion (BIC) values for each category. Smaller BIC values indicated more convincing models for predicting results.(13)

Stratum analysis and univariate COX proportional hazards Analysis were used to assess the impact of the clinicopathological characteristics on the efficiency of prognostic prediction of the MLNR for patients. All statistical analyses were performed using IBM SPSS Statistics (Version 22.0; IBM Corp. New York, USA). And p values < 0.05 were considered statistically significant.

Results

General results

A total of 3864 patients undergoing curative gastrectomy with nodal dissection were eligible for this study between February 2011 and February 2016. The 5-year survival rate (YSR) of 3864 GC patients was 54.7%, with a median overall survival (OS) of 63.03 months (range: 1.13-110.33 months) (Figure S2).

2872 (5-YSR: 55.1%) patients were male and 992 (5-YSR: 53.6%) were female, with a median age of 63 years (range: 19–90 years). 2570 (5-YSR: 52.8%) patients were aged 60 years or older while 1294 (5-YSR: 58.6%) were less than 60 years old. Regarding the tumor location, there were 1860 (5-YSR: 53.5%), 606 (5-YSR: 52.8%), 1104 (5-YSR: 62.5%), and 294 (5-YSR: 36.7%) tumors in the lower third, middle third, upper third, and > 2/3 stomach groups, respectively.

In view of the type of gastrectomy, there were 2628 (5-YSR: 49.5%), 1048 (5-YSR: 65.6%), and 188 (5-YSR: 66.0%) patients in the total gastrectomy (TG), distal gastrectomy (DG), proximal gastrectomy (PG) groups, respectively. Considering perineural invasion, there were 390 (5-YSR: 43.1%), and 3474 (5-YSR: 56.0%) patients in the positive, and negative groups, respectively. With respect to vascular invasion, there were 642 (5-YSR: 39.6%), and 3222 (5-YSR: 57.1%) patients in the positive, and negative groups, respectively.

In addition, of 3864 patients, 608 (5-YSR: 89.5%) were staged as having pT1 tumors, 326 (5-YSR: 77.9%) as pT2, 254 (5-YSR: 61.4%) as pT3, 2124 (5-YSR: 45.3%) as pT4a, and 552 (5-YSR: 35.9%) as pT4b. Patients were divided into five groups based on N stage (N0, N1, N2, N3a, and N3b), there were 1476 (5-YSR: 80.1%), 640 (5-YSR: 58.6%), 796 (5-YSR: 41.7%), 753 (5-YSR: 27.5%) and 200 (5-YSR: 9.0%) patients in each group respectively. Also, patients were divided into three groups based on TNM stage (I, II, III), there were 784 (5-YSR: 90.1%), 834 (5-YSR: 70.9%), and 2246 (5-YSR: 36.3%) patients in each group respectively.

The most appropriate cut-off value for MLNR was 0.25 ($X^2 = 866.433$, $P < 0.001$) based on the "minimum p-value" approach by the X-tile software (Fig. 1). Of 3864 patients, 2548 (5-YSR: 69.5%) were classified into a low-MLNR (L-MLNR) group, and 1316 (5-YSR: 26.1%) were classified into a high-MLNR (H-MLNR) group. The most appropriate cut-off value for tumor size was 4.5 cm ($X^2 = 454.250$, $P < 0.001$) (Figure S1). Patients were divided into two groups based on tumor size (< 4.5 cm and ≥ 4.5 cm), and there were 1872 (5-YSR: 71.8%), and 1948 (5-YSR: 38.4%) patients in each group respectively. The data of the included 3864 patients were analyzed, with the clinicopathological characteristics of the patients shown in Table 1.

Table 1
Clinicopathological characteristics and survival analyses of GC patients (N = 3864)

Characteristics	n	5-YSR (%)	χ^2	Univariate P value	HR (95% CI)	Multivariate P value
Gender			0.014	0.906		0.699
Male	2872	55.1				
Female	992	53.6				
Age at surgery (year)			15.521	< 0.001		< 0.001
< 60	1294	58.6			Reference	
≥ 60	2570	52.8			1.242 (1.123–1.375)	
Tumor location			77.078	< 0.001		0.447
Upper third	1860	53.5				
Middle third	606	52.8				
Lower third	1104	62.5				
> 2/3 stomach	294	36.7				
Tumor size (cm)*			457.908	< 0.001		< 0.001
<4.5	1872	71.8			Reference	
≥4.5	1948	38.4			1.599 (1.437–1.781)	
Type of gastrectomy			95.569	< 0.001		< 0.001
TG	2628	49.5			Reference	Reference
DG	1048	65.6			0.745 (0.662–0.838)	< 0.001
PG	188	66.0			0.827 (0.646–1.060)	0.133
T stage			488.722	< 0.001		0.302

GC, gastric cancer; TG, total gastrectomy; DG, distal gastrectomy; PG, proximal gastrectomy; ELN, examined lymph node; 5-YSR, 5-year survival rate; HR, hazard ratio; 95% CI, 95% confidence interval; *, median of tumor diameter: 4.5 (0.3 – 27.0) cm; MLNR: metastatic lymph

node ratio; The eighth edition of TNM classification for GC was adopted for postoperatively pathological stages of all included patients

Characteristics	n	5-YSR (%)	χ^2	Univariate P value	HR (95% CI)	Multivariate P value
T1	608	89.5				
T2	326	77.9				
T3	254	61.4				
T4a	2124	45.3				
T4b	552	35.9				
N stage			1105.510	< 0.001		< 0.001
N0	1476	80.1			0.375 (0.271–0.518)	< 0.001
N1	640	58.6			0.408 (0.311–0.535)	< 0.001
N2	796	41.7			0.497 (0.406–0.609)	< 0.001
N3a	752	27.5			0.566 (0.476–0.673)	< 0.001
N3b	200	9.0			Reference	Reference
ELN			1.268	0.260		< 0.001
<16	1238	56.9			Reference	
≥16	2626	53.7			0.807 (0.726–0.897)	
Perineural invasion			6.271	0.012		0.023
Positive	390	43.1			Reference	
Negative	3474	56.0			1.187 (1.024–1.376)	
Vascular invasion			100.000	< 0.001		0.005
Positive	642	39.6			Reference	

GC, gastric cancer; TG, total gastrectomy; DG, distal gastrectomy; PG, proximal gastrectomy; ELN, examined lymph node; 5-YSR, 5-year survival rate; HR, hazard ratio; 95% CI, 95% confidence interval; *, median of tumor diameter: 4.5 (0.3 – 27.0) cm; MLNR: metastatic lymph

node ratio; The eighth edition of TNM classification for GC was adopted for postoperatively pathological stages of all included patients

Characteristics	n	5-YSR (%)	χ^2	Univariate P value	HR (95% CI)	Multivariate P value
Negative	3222	57.7			0.841 (0.746–0.949)	
TNM stage			788.209	< 0.001		< 0.001
I	784	90.1			Reference	Reference
II	834	70.9			2.237 (1.753–2.855)	< 0.001
III	2246	36.3			3.642 (2.714–4.888)	< 0.001
MLNR			883.883	< 0.001		< 0.001
< 0.25	2548	69.5			Reference	
≥ 0.25	1316	26.1			1.526 (1.279–1.821)	
GC, gastric cancer; TG, total gastrectomy; DG, distal gastrectomy; PG, proximal gastrectomy; ELN, examined lymph node; 5-YSR, 5-year survival rate; HR, hazard ratio; 95% CI, 95% confidence interval; *, median of tumor diameter: 4.5 (0.3 – 27.0) cm; MLNR: metastatic lymph						
node ratio; The eighth edition of TNM classification for GC was adopted for postoperatively pathological stages of all included patients						

Survival analysis

The median follow-up time was 62.7 (range: 1.4 – 110.2) months. Clinicopathological factors were analyzed to verify prognostic significance in terms of OS in patients. Univariable analysis demonstrated that age at surgery ($P < 0.001$), tumor location ($P < 0.001$), tumor size ($P < 0.001$), type of gastrectomy ($P < 0.001$), T stage ($P < 0.001$), N stage ($P < 0.001$), perineural invasion ($P = 0.012$), vascular invasion ($P < 0.001$), TNM stage ($P < 0.001$), and MLNR ($P < 0.001$) were significant predictors of OS (Fig. 2,3).

Significant differences were observed among the low and high-MLNR groups ($P < 0.001$); the 5-YSRs were 69.5%, and 26.1%, respectively. Multivariable analyses demonstrated that age at surgery ($P < 0.001$), tumor size ($P < 0.001$), type of gastrectomy ($P < 0.001$), N stage ($P < 0.001$), ELN ($P < 0.001$), vascular invasion ($P = 0.005$), TNM stage ($P < 0.001$), and MLNR ($P < 0.001$) were significant independent predictors of OS (Table 1).

Relationship between clinicopathological characteristics and MLNR of GC patients

Of 3864 GC patients, the Chi-square test analysis demonstrated that tumor location ($P < 0.001$), tumor size ($P < 0.001$), type of gastrectomy ($P < 0.001$), T stage ($P < 0.001$), N stage ($P < 0.001$), perineural

invasion ($P < 0.001$), vascular invasion ($P < 0.001$), and TNM stage ($P < 0.001$) were significantly related to the MLNR.

Multivariable analyses demonstrated that age at surgery ($P = 0.030$, BIC: 876), tumor location ($P = 0.005$, BIC: 868), N stage ($P < 0.001$, BIC: 1916) and ELN ($P < 0.001$, BIC: 1023) were significant independent factors of MLNR. In addition, the multinomial logistic regression model showed the smallest BIC values for age at surgery and tumor location, indicating the most intensive relationship with MLNR (Table 2). The linear correlation between the ELN count and the MLNR was illustrated in Supplementary Fig. 3 ($R^2 = 0.0008$, $P = 0.0778$).

Table 2

Relationship between clinicopathological characteristics and Metastatic Lymph Node Ratio (n = **3864**)

Characteristics	Metastatic Lymph Node Ratio		Univariable analysis		Multivariable analysis		
	< 0.25	≥ 0.25	χ^2	P value	OR (95%CI)	P value	BIC value
Gender			0.357	0.550		0.316	
Male	1906	966					
Female	648	344					
Age at surgery (year)			3.319	0.068		0.030	876
< 60	830	464			Reference		
≥ 60	1724	846			0.702 (0.511–0.966)		
Tumor location			46.407	< 0.001		0.005	868
Upper third	1308	552			Reference	Reference	
Middle third	358	248			1.785 (1.184–2.693)	0.006	
Lower third	728	376			0.915 (0.639–1.309)	0.627	
> 2/3 stomach	160	134			0.579 (0.293–1.147)	0.117	
Tumor size (cm)			276.692	< 0.001		0.402	
<4.5	1482	390					
≥4.5	1046	902					
Type of gastrectomy			17.698	< 0.001		0.060	
TG	1690	938					

BIC: Bayesian information criterion; TG, total gastrectomy; DG, distal gastrectomy; PG, proximal gastrectomy; ELN, examined lymph node; OR, odds ratio; 95% CI, 95% confidence interval; The depth of primary tumor invasion (T stage), classification of regional metastasis lymph nodes (N stage) and TNM stage were based on the 8th edition TNM staging system.

Characteristics	Metastatic Lymph Node Ratio		Univariable analysis		Multivariable analysis	
DG	718	330				
PG	146	42				
T stage			462.895	< 0.001	0.334	
T1	590	18				
T2	278	48				
T3	174	80				
T4a	1262	862				
T4b	250	302				
N stage			2888.807	< 0.001	< 0.001	1916
N0	1476	0				
N1	628	12				
N2	434	362				
N3a	10	742				
N3b	0	200				
ELN			2.041	0.153	< 0.001	1023
≤16	836	402			Reference	
≥16	1712	914			0.129 (0.091–0.182)	
Perineural invasion			81.004	< 0.001	0.719	
Positive	178	212				
Negative	2376	1098				
Vascular invasion			198.583	< 0.001	0.914	
Positive	270	372				
BIC: Bayesian information criterion; TG, total gastrectomy; DG, distal gastrectomy; PG, proximal gastrectomy; ELN, examined lymph node; OR, odds ratio; 95% CI, 95% confidence interval; The depth of primary tumor invasion (T stage), classification of regional metastasis lymph nodes (N stage) and TNM stage were based on the 8th edition TNM staging system.						

Characteristics	Metastatic Lymph Node Ratio		Univariable analysis		Multivariable analysis
Negative	2284	938			
TNM stage			1240.679	< 0.001	0.471
I	784	0			
II	794	40			
III	970	1276			
BIC: Bayesian information criterion; TG, total gastrectomy; DG, distal gastrectomy; PG, proximal gastrectomy; ELN, examined lymph node; OR, odds ratio; 95% CI, 95% confidence interval; The depth of primary tumor invasion (T stage), classification of regional metastasis lymph nodes (N stage) and TNM stage were based on the 8th edition TNM staging system.					

3.4 Effects of other clinicopathological characteristics on MLNR for predicting prognosis of GC patients

Stratum analysis was used to assess the impact of the clinicopathological characteristics on the efficiency of prognostic prediction of the MLNR for GC patients.

We validated four clinicopathological factors (age at surgery, tumor location, N stage and ELN) that were statistically significant for MLNR in Table 3.

Table 3
Effects of clinicopathological characteristics on Metastatic Lymph Node Ratio for predicting prognosis of GC patients

Characteristics	MLNR	No. of patients (%)	5-YSR (%)	χ ²	P value
Age at surgery (year)					
< 60	< 0.25	830	616 (74.2)	300.799	< 0.001
	≥ 0.25	464	142 (30.6)		
≥ 60	< 0.25	1724	1154 (66.9)	586.187	< 0.001
	≥ 0.25	846	202 (23.9)		
Tumor location					
Upper third	< 0.25	1308	858 (65.6)	359.092	< 0.001
	≥ 0.25	552	138 (25.0)		
Middle third	< 0.25	358	246 (68.7)	123.769	< 0.001
	≥ 0.25	248	74 (29.8)		
Lower third	< 0.25	728	572 (78.6)	295.891	< 0.001
	≥ 0.25	376	118 (31.4)		
> 2/3 stomach	< 0.25	160	94 (58.8)	82.958	< 0.001
	≥ 0.25	134	14 (10.4)		
N stage					
N0	< 0.25	1476	1182 (80.1)	-	-
	≥ 0.25	0			
N1	< 0.25	628	372 (59.2)	6.712	0.010
	≥ 0.25	12	4 (33.3)		
N2	< 0.25	434	206 (47.5)	17.955	< 0.001
	≥ 0.25	362	126 (34.8)		
N3a	< 0.25	10	10 (100.0)	13.552	< 0.001
	≥ 0.25	742	196 (26.4)		
N3b	< 0.25	0		-	-
	≥ 0.25	200	18 (9.0)		

MLNR: metastatic lymph node ratio; ELN, examined lymph node; 5-YSR, 5-year survival rate

Characteristics	MLNR	No. of patients (%)	5-YSR (%)	χ^2	P value
ELN					
<16	< 0.25	836	592 (70.8)	282.830	< 0.001
	≥ 0.25	402	112 (27.9)		
≥ 16	< 0.25	1712	1178 (68.8)	599.797	< 0.001
	≥ 0.25	914	232 (25.4)		
MLNR: metastatic lymph node ratio; ELN, examined lymph node; 5-YSR, 5-year survival rate					

Table 4
MLNR predicting prognosis of GC patients (univariate Cox regression)

Characteristics	MLNR < 0.25		MLNR ≥ 0.25	
	HR (95% CI)	P	HR (95% CI)	P
Gender	Reference	Reference	0.269 (0.245–0.295)	< 0.001
Age at surgery	Reference	Reference	0.266 (0.242–0.292)	< 0.001
Tumor location	Reference	Reference	0.271 (0.247–0.297)	< 0.001
Tumor size	Reference	Reference	0.335 (0.304–0.369)	< 0.001
Type of gastrectomy	Reference	Reference	0.274 (0.249–0.300)	< 0.001
T stage	Reference	Reference	0.362 (0.328–0.398)	< 0.001
N stage	Reference	Reference	0.625 (0.528–0.740)	< 0.001
ELN	Reference	Reference	0.269 (0.245–0.295)	< 0.001
Perineural invasion	Reference	Reference	0.270 (0.245–0.296)	< 0.001
Vascular invasion	Reference	Reference	0.278 (0.253–0.306)	< 0.001
TNM stage	Reference	Reference	0.478 (0.431–0.531)	< 0.001
GC, gastric cancer; ELN, examined lymph node; HR, hazard ratio; 95% CI, 95% confidence interval; MLNR: metastatic lymph node ratio; The depth of primary tumor invasion (T stage), classification of regional metastasis lymph nodes (N stage) and TNM stage were based on the 8th edition TNM staging system.				

Firstly, we have evaluated the influence of MLNR on prognosis in both group 1 (age < 60) and group 2 (age ≥ 60). In group 1, there were 1294 patients, who were divided into L-MLNR and H-MLNR ($P < 0.001$) (Fig. 4A); the 5-YSRs were 74.2%, and 30.6%, respectively. In group 2, there were 2626 patients, who were divided into MLNR < 0.25 and MLNR ≥ 0.25 ($P < 0.001$) (Fig. 4B); the 5-YSRs were 66.9%, and 23.9%, respectively.

Secondly, we have evaluated the influence of MLNR on prognosis in group 1 (Upper third), group 2 (Middle third), group 3 (Lower third) and group 4 (> 2/3 stomach). In group 1, there were 1860 patients, who were divided into $MLNR < 0.25$ and $MLNR \geq 0.25$ ($P < 0.001$) (Fig. 4E); the 5-YSRs were 65.6%, and 25.0%, respectively. In group 2, there were 606 patients, who were divided into $MLNR < 0.25$ and $MLNR \geq 0.25$ ($P < 0.001$) (Fig. 4F); the 5-YSRs were 68.7%, and 29.8%, respectively. In group 3, there were 1104 patients, who were divided into $MLNR < 0.25$ and $MLNR \geq 0.25$ ($P < 0.001$) (Fig. 4G); the 5-YSRs were 78.6%, and 31.4%, respectively. In group 4, there were 294 patients, who were divided into $MLNR < 0.25$ and $MLNR \geq 0.25$ ($P < 0.001$) (Fig. 4H); the 5-YSRs were 58.8%, and 10.4%, respectively.

Thirdly, we have evaluated the influence of MLNR on prognosis in group 1 (N0), group 2 (N1), group 3 (N2), group 4 (N3a), and group 5 (N3b). In group 1, there were 1476 patients, who were $MLNR < 0.25$; the 5-YSRs were 80.1%. In group 2, there were 640 patients, who were divided into $MLNR < 0.25$ and $MLNR \geq 0.25$ ($P < 0.001$) (Fig. 5B); the 5-YSRs were 59.2%, and 33.3%, respectively. In group 3, there were 796 patients, who were divided into $MLNR < 0.25$ and $MLNR \geq 0.25$ ($P < 0.001$) (Fig. 5C); the 5-YSRs were 47.5%, and 34.8%, respectively. In group 4, there were 752 patients, who were divided into $MLNR < 0.25$ and $MLNR \geq 0.25$ ($P < 0.001$) (Fig. 5D); the 5-YSRs were 100.0%, and 26.4%, respectively. In group 5, there were 200 patients, who were $MLNR \geq 0.25$; the 5-YSRs were 9.0%.

Finally, we have evaluated the influence of MLNR on prognosis in group 1 ($ELN < 16$) and group 2 ($ELN \geq 16$). In group 1, there were 1238 patients, who were divided into $MLNR < 0.25$ and $MLNR \geq 0.25$ ($P < 0.001$) (Fig. 4C); the 5-YSRs were 70.8%, and 27.9%, respectively. In group 2, there were 2570 patients, who were divided into $MLNR < 0.25$ and $MLNR \geq 0.25$ ($P < 0.001$) (Fig. 4D); the 5-YSRs were 68.8%, and 25.4%, respectively. The results showed that MLNRs were statistically significant at all levels of clinicopathological factors.

To make the above results more convincing, we used the univariate COX proportional hazards analysis to assess the impact of MLNR on the prognosis of GC patients. We discovered that MLNR had a significant impact on prognosis in distinguishing substages of patients with clinicopathological features, such as gender [$P < 0.001$, 0.269 (0.245–0.295)], age at surgery [$P < 0.001$, 0.266 (0.242–0.292)], tumor location [$P < 0.001$, 0.271 (0.247–0.297)], tumor size [$P < 0.001$, 0.335 (0.304–0.369)], type of gastrectomy [$P < 0.001$, 0.274 (0.249–0.300)], T stage [$P < 0.001$, 0.362 (0.328–0.398)], N stage [$P < 0.001$, 0.625 (0.528–0.740)], ELN [$P < 0.001$, 0.269 (0.245–0.295)], perineural invasion [$P < 0.001$, 0.270 (0.245–0.296)], vascular invasion [$P < 0.001$, 0.278 (0.253–0.306)], and TNM stage [$P < 0.001$, 0.478 (0.431–0.531)].

Discussion

Gastric cancer is one of the most common malignancies in the world, causing thousands of deaths each year, especially in China.(14, 15) The possibility of prolonged survival after curative resection of GC has been explored by researchers worldwide.(16)

Therefore, it is widely studied that the factors affecting the prognosis after curative resection of gastric cancer.(17) Indisputably, TNM staging has important value in assessing the prognosis of gastric cancer

patients.(18) Although the American Joint Commission for Cancer (AJCC) has recommended an ELN ≥ 16 since 2009 to ensure accurate prognosis at N stage, the 8th edition of the TNM classification for gastric cancer does not mandate a minimum ELN count required for correct staging.(19) In 2018, Deng, J. et al. conducted a study exploring the impact of ELN count on enhancing the accuracy of prognostic evaluation of GC patients after curative gastrectomy. The study indicated that the 5-YSR of GC patients with ELNs ≥ 16 was significantly superior than that of patients with ELNs < 16 ($P < 0.001$). (19) There were similar results in our study, multivariable analyses demonstrated that ELNs < 16 has a worse OS than ELNs ≥ 16 [0.807 (0.726–0.897), $P < 0.001$].

Some studies showed that most GC patients in China were advanced cases at the time of initial diagnosis, and theoretically staging migration may occur in Chinese GC patients with insufficient ELN counts.(20–22) In 2019, Dong, Y. et al. conducted a study to validate the stage migration related to the insufficient ELN count in pN3a GC patients. The results demonstrated that stage migration was predominantly detected in pN3a patient with insufficient (less than 16) ELN count. (21) Therefore, detection of additional lymph nodes from postoperative specimens is considered necessary and useful. (23) Although most surgeons follow the UICC/AJCC guidelines and remove a sufficient number of lymph nodes, there are still possibilities of examining fewer than 16 lymph nodes on account of lack of surgical experience, small lymph nodes in the sample, and inadequacy of the technique itself.(24–27) An analysis of 18,043 GC patients from the Surveillance, Epidemiology, and End Results Program (SEER) database, studies from 18 hospitals in England and 691 hospitals in the USA found that only 33%, 31% and 40% of their respective gastrectomy met the minimum recommended number of LNs for surgical retrieval.(28) So "stage migration" is a common and misleading phenomenon in TNM staging, in 10–25% of cases.(29)

It is of great importance for us to find a factor independent of TNM classification providing a better reference of prognosis of GC patients.(30) Recently, several researchers have demonstrated that MLNR was of prominent benefit to guide in clinical diagnosis in many cancers, including breast, colon and rectal cancer.(6, 7) In addition, some investigators have proposed that MLNR could be a simple and useful prognostic predictor in patients with GC. (10, 31, 32) In 2018, Hou, Y. et al. conducted a retrospective study included 221 patients who got GC and underwent curative gastrectomy. The results indicated that MLNR could estimate the prognosis of patients with curative gastrectomy. (32) In 2020, Kano, K. et al. conducted a retrospective study included 838 patients with Pathological Stage II/III GC who underwent curative gastrectomy. The results indicated that High MLNR is a predictor of worse prognosis in Pathological Stage II/III GC. (10)

The purpose of this paper was to determine the prognostic significance of MLNR to assess the prognosis of patients undergoing curative gastrectomy. A larger sample size was included in this study. In this study, a total of 3864 patients were divided into two groups based on the most appropriate cut-off value of MLNR. The optimal cutoff value for MLNR was defined as 0.25 by using X-tile software. The prognosis of patients who had an L-MLNR was significantly better than that of patients who had an H-MLNR. We found that age at surgery, tumor location, tumor size, type of gastrectomy, T stage, N stage, peripheral nerve invasion, vascular invasion, TNM stage, and MLNR were distinct factors affecting patient prognosis

based on univariate analysis. Furthermore, using the Cox proportional-hazards model, a multivariable analysis, we also observed a similar finding that all of the ten factors contributed to prognosis improvement in GC patients.

Our study demonstrated that MLNR was a significant independent predictor of OS via both univariate and multivariate analyses, which meant that GC patients with a lesser MLNR would have a longer survival time. Therefore, this investigation would shed light on a promising perspective that MLNR could be a new focus to estimate the prognosis of patients undergoing curative gastrectomy. To make it more reliable and convincing, we further investigated the relationship between clinicopathological features and MLNR of GC patients. Both univariable analysis and multivariable analyses indicated that age at surgery, tumor location, N stage and ELN were significant independent factors of MLNR. Additionally, we performed stratum analysis and univariate COX proportional hazards analysis to validate the influences of MLNR on prognosis in various patient subgroups with diverse clinicopathological characteristics.

Our results showed that in the ELN group, MLNR was statistically significant in predicting prognosis for $ELN < 16$ and $ELN \geq 16$. In other words, according to our findings, MLNR was a prognostic indicator for GC patients, regardless of whether the number of lymph nodes examined is < 16 or ≥ 16 . Besides, the median survival time decreased with increasing MLNR. In addition, we found no correlations between MLNR and the total count of ELNs ($R^2 = 0.0008$, $P = 0.0778$). This was similar to the results of Hou, Y. et al that there was no correlation between the MLNR and the ELNs.(32) Patients with the same MLNR would have similar results even if the number of detected metastatic nodes was different. On the contrary, patients with higher MLNR would have an unfavorable outcome even though they had the same count of metastatic nodes. Therefore, MLNR may be a new prognostic indicator to complement the TNM staging system.

Several limitations should be noted in our research. First, this was a retrospective single-center study. We expect to carry out prospective and multi-center investigations in the future. Second, all patients included were from the Chinese population, which may lead to a slight bias in the test results relative to other races. As approximately 42% of global GC patients occur in China, it is worthwhile for us to conduct cross-ethnic study for comprehensive guiding of MLNR and optimal clinical therapies. Third, neoadjuvant chemotherapy has recently become a standard therapy regimen, especially for high-risk GCs. However, it is unclear whether LNR can be a prognostic factor for patients undergoing radical resection after neoadjuvant chemotherapy. Considering these limitations, the results should be further validated in future prospective multicenter studies.

Conclusions

In conclusion, MLNR is a significant independent predictor for assessing the prognosis of patients undergoing curative gastrectomy, including the individual substages of clinicopathological features (gender, age at surgery, tumor location, tumor size, type of gastrectomy, T stage, N stage, ELN, perineural

invasion, vascular invasion, and TNM stage). Therefore, MLNR may become a new indicator for evaluating prognosis of patients with curative gastrectomy and enhance the current guidelines.

Abbreviations

AJCC, American Joint Commission for Cancer; BIC, Bayesian information criterion;

DG, distal gastrectomy; ELN, examined lymph node; GC, gastric cancer; HR, hazard ratio; UICC, The Union for International Cancer Control; MLNR: metastatic lymph node ratio; OS, Overall survival; PG, proximal gastrectomy; SEER, Surveillance, Epidemiology, and End Results; TG, total gastrectomy; TNM, Tumor–node–metastasis; 5-YSR, 5-year survival rate; 95% CI, 95% confidence interval.

Declarations

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None.

Author contributions

Conceptualization of the study was by Z He, K Chen and Z Zhang. Methodology was devised by Z He. Writing, review and editing were performed by Z He and D Li, and the manuscript was revised by Z He, S Liu and K Chen. Data collation was done by D Li, Y Lu, E Song and S Liu. Z Zhang was responsible for project administration.

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Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request. All data generated or analyzed during this study are included in this published article [and its supplementary information files].

Disclosure Statement

The authors have no other relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter discussed in the manuscript apart from those disclosed.

Ethics approval and consent to participate

All procedures followed were in accordance with the ethical standards of The First Affiliated Hospital of Anhui Medical University ethics committee and with the Helsinki Declaration of 1964 and later versions. An informed written and verbal consent was obtained from all the studied patients.

Consent for publication

All authors declare that a written and verbal informed consent was obtained from the selected participants in the study for publication of the data.

Competing interests

The authors declare no conflict of interest.

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Figures

Figure 1

X-tile plots of MNLR on gastric cancer following curative resection. The X-tile plots show the χ^2 log-rank values with cut points, producing low and high subgroups. A. The X-axis represents all potential cutoffs from low to high (left to right), defining a low subset, while the Y-axis represents cutoffs from high to low (top to bottom), defining a high subset. The red color of the cutoff values indicates an inverse relationship with the recurrence time, while green color indicates a direct correlation. Best cutoff value appears in the brightest pixel (green or red). B. The entire cohort was divided into low and high histograms based on the optimal cutoff value of MLNR (0.25). C. A Kaplan-Meier plot of overall survival of gastric cancer patients generated from the optimal cutoff value of MLNR. Blue represents the low subgroup and gray represents the high subgroup.

Figure 2

The factors with statistical significance of univariate analysis and panels A to H reveal the survival curves of age at surgery, tumor location, tumor size, type of gastrectomy, T stage, N stage, perineural invasion, and vascular invasion, respectively.

Figure 3

The factors with statistical significance of univariate analysis and panels A to B reveal the survival curves of TNM stage, and MLNR respectively.

Figure 4

The survival curves of MLNR in group 1 (Age < 60), group 2 (Age ≥ 60), group 3 (ELN < 16), group 4 (ELN ≥ 16), group 5 (Upper third), group 6 (Middle third), group 7 (Lower third), and group 8 (>2/3 stomach), respectively. Panels A and H reveal the survival curves of MLNR in group 1 (Age < 60), group 2 (Age ≥ 60), group 3 (ELN < 16), group 4 (ELN ≥ 16), group 5 (Upper third), group 6 (Middle third), group 7 (Lower third), and group 8 (>2/3 stomach), respectively.

Figure 5

The survival curves of MLNR in group 1 (N0), group 2 (N1), group 3 (N2), group 4 (N3a), and group 5 (N3b), respectively. Panels A and E reveal the survival curves of MLNR in group 1 (N0), group 2 (N1), group 3 (N2), group 4 (N3a), and group 5 (N3b), respectively.

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