Received: 17 February 2020 | Revised: 22 April 2020 | Accepted: 11 May 2020

DOI: 10.1111/jop.13043

ORIGINAL ARTICLE

Journal of Oral Pathology & Medicine WILEY

High-risk lymph node ratio predicts worse prognosis in patients with locally advanced oral cancer

Tong-Chao Zhao[1] | Si-Yuan Liang[1] | Wu-Tong Ju[1] | Yong Fu[1] | Zhi-Hang Zhou[1] | Li-Zhen Wang[2] | Jiang Li[2] | Chen-Ping Zhang[1] | Zhi-Yuan Zhang[1] | Lai-Ping Zhong[1]

[1]Department of Oral & Maxillofacial-Head & Neck Oncology, Ninth People's Hospital, Shanghai Jiao Tong University School of Medicine, Shanghai, China

[2]Department of Oral Pathology, Ninth People's Hospital, Shanghai Jiao Tong University School of Medicine, Shanghai, China

Correspondence
Lai-Ping Zhong, Department of Oral & Maxillofacial-Head & Neck Oncology, Ninth People's Hospital, Shanghai Jiao Tong University School of Medicine, No 639, Zhizaoju Rd, Shanghai 200011, China.
Email: zhonglp@hotmail.com

Wu-Tong Ju, Department of Oral & Maxillofacial-Head & Neck Oncology, Ninth People's Hospital, Shanghai Jiao Tong University School of Medicine, No 639, Zhizaoju Rd, Shanghai 200011, China.
Email: juwt9th@163.com

Funding information
National Natural Science Foundation of China, Grant/Award Number: 81672660; Clinical Research Project of Multi-Disciplinary Team, Ninth People's Hospital, Shanghai Jiao Tong University School of Medicine, Grant/Award Number: 201916; Shanghai Municipal Commission of Health and Family Planning, Grant/Award Number: 2018BR41; Two-hundred Talent, Shanghai Jiao Tong University School of Medicine, Grant/Award Number: 20191915

Abstract

Background: To investigate the prognostic value of lymph node ratio (LNR), as well as the correlation with docetaxel, cisplatin, and 5-FU (TPF) induction chemotherapy, in patients with locally advanced oral squamous cell carcinoma (OSCC).

Methods: Two-hundred and forty-five patients from a phase 3 trial involving TPF induction chemotherapy in stage III/IVA OSCC patients (NCT01542931) were enrolled in this study between 2008 and 2010. The clinical and pathological data were collected and analyzed. The cutoff value for LNR was calculated on the receiver operating characteristic (ROC) curve. Univariate and multivariate Cox regression models, and Kaplan-Meier method were used for survival analysis.

Results: According to the ROC curve, the cutoff value for LNR was 7.6%. With a median follow-up period of 80 months, the OSCC patients with high-risk LNR (> 7.6%), or positive extranodal extension (ENE) had significantly worse clinical outcomes than patients with low-risk LNR (≤7.6%) or negative ENE. Multivariate analysis on pathological covariates showed that only high-risk LNR was an independent negative predictive factor for survival ($P < .05$). The cutoff value of LNR of 7.6% was also verified with the similar results using an open TCGA database, high-risk LNR indicating worse overall survival ($P < .001$) and disease-free survival ($P < .001$).

Conclusion: Oral squamous cell carcinoma patients with high-risk LNR have a worse clinical outcome than patients with low-risk LNR. High-risk LNR is an independent negative predictive factor for clinical outcome in patients with locally advanced OSCC.

KEYWORDS
lymph node ratio, oral cancer, prognosis, TPF induction chemotherapy

Tong-Chao Zhao and Si-Yuan Liang contributed equally to this study.

The peer review history for this article is available at https://publons.com/publon/10.1111/jop.13043

1 | INTRODUCTION

Lymph node status plays a vital role in predicting survival outcomes in patients with oral cancer. Pathological nodal (pN) stage is a very important indicator for treatment plan and clinical outcomes in patients with oral squamous cell carcinoma (OSCC). It is generally classified by the number of positive nodes and/or location of the most advanced pathological node excised, which mainly depends on the surgical and pathological procedures. Given that an inadequate number of nodes excised, as well as differences in surgical and sampling skills between institutions or hospitals may affect the pN stage, the lymph node ratio (LNR) is introduced in an attempt to reduce the potential bias by utilizing both the disease regional extension (number of positive lymph nodes [pLNN]) and the surgical and sampling differences (total number of lymph nodes [LNN]), which are excised and examined from the neck dissection). LNR, calculated by the formula, LNR = pLNN/LNN, has received increased attention as a reliable prognostic factor in various tumors.[1-4] Several studies have reported that LNR is a better predictor than the conventional pN stage in OSCC patients, thus indicating that LNR is an independent prognostic factor for OSCC patients.[4-9]

We previously conducted a phase 3 trial involving docetaxel, cisplatin, and 5-FU (TPF) induction chemotherapy in patients with locally advanced and resectable OSCC (registration ID, NCT01542931).[10] Neither short- nor long-term follow-up results from the initial trial revealed a significant survival benefit of TPF induction chemotherapy with respect to clinical outcome. Indeed, only a proportion of pathological responders benefitted significantly from induction chemotherapy. Subgroup analysis showed that the patients with clinical N2 stage also benefitted from TPF induction chemotherapy.[10,11] To date, there have been no studies analyzing the correlation between LNR and treatment protocol including TPF induction chemotherapy in OSCC patients. LNR has not been reported in the latest National Comprehensive Cancer Network (NCCN) Clinical Practice Guidelines in head and neck cancers.

Based on the clinical and pathological data from the phase 3 trial (NCT01542931), in the current study, we determined the relationship between LNR and TPF induction chemotherapy in patients with locally advanced OSCC, especially the clinical outcome.

2 | METHODS

2.1 | Patients

This study was retrospective, from March 2008 to December 2010, and two-hundred and fifty-six OSCC patients (179 males and 77 females; age range, 26-75 years; mean age, 55.4 years) at clinical stages III and IVA were enrolled in a previous phase 3 trial (NCT01542931) that determined the potential benefit of TPF induction chemotherapy prior to standard treatment in locally advanced OSCC patients. All of the patients were from the Department of Oral & Maxillofacial-Head & Neck Oncology at the Ninth Peoples' Hospital, Shanghai Jiao Tong University School of Medicine (Shanghai, China). The detailed protocol of the trial has been previously described.[10] Briefly, patients who met the criteria were randomly assigned to the experimental (TPF induction chemotherapy + surgery +postoperative radiotherapy) or control group (surgery + postoperative radiotherapy); there were 128 patients in each group. Among the 256 patients, six patients declined surgical treatment, two patients died as a result of traffic accidents during the period of induction chemotherapy. 248 patients with surgical treatment information were enrolled in this study, their pathological data (especially the lymph nodes) and clinical data were collected; the pathological data were used for detail analysis. Twenty-two patients (ten in the experimental group and twelve in the control group) refused the postoperative radiotherapy or died before the radiotherapy. This study was approved by the Ethics committee of Ninth People's Hospital, Shanghai Jiao Tong University School of Medicine. All the participants gave written informed consent.

2.2 | Lymph node evaluation

According to the trial protocol, the patients underwent neck dissection during the surgery. The neck dissection specimens were separated by the surgical team and assessed by oral pathologists according to the guidelines.[12,13] All lymph nodes separated from neck dissection, which could be identified larger than 3 mm in size, were fixed in 10% neutralized formalin and embedded in paraffin for use. Each lymph node was cut into sections with 2-3 mm intervals along the long axis, and one slice from each section was used for HE staining and analysis. After evaluated by two pathologists, the patient was identified as pN0 when no tumor cells found in all lymph nodes from neck dissection. The positive metastatic lymph node was measured, and the extranodal extension (ENE) status was recorded.[14,15] The number of total nodes, positive nodes, and ENE status was collected and recorded.

2.3 | Follow-up

Patients were followed up every 3 months for the first 2 years after treatment, every 6 months for the next 3-5 years, and every year after 5 years until death or data censoring. Patient survival time was counted from the date of random assignment. The event of overall survival (OS) was defined as the occurrence of death of any cause in the patients. The event of disease-free survival (DFS) was tumor recurrence or the occurrence of death of any cause. The event of disease-specific survival (DSS) was the occurrence of death of OSCC. The event of locoregional recurrence-free survival (LRFS) was local tumor or neck recurrence or the occurrence of death of any cause. The event of distant metastasis-free survival (DMFS) was tumor distant metastasis or occurrence of death of any cause.

TABLE 1 Summary of pathological characteristics of 248 patients

	Total		LNR ≤ 0.076		LNR > 0.076		
	(N = 248)		(N = 181)		(N = 67)		
Variable	N	%	N	%	N	%	P^*
Pathological T stage							
T0	15	6.1	12	6.7	3	4.5	.537
T1	35	14.1	25	13.8	10	14.9	
T2	71	28.6	47	26.0	24	35.8	
T3	90	36.3	70	38.7	20	29.9	
T4	37	14.9	27	14.9	10	14.9	
Pathological N stage							
N0	103	41.5	103	56.9	0	0	**<.001**
N1	41	16.5	36	19.9	5	7.5	
N2	104	42.0	42	23.2	62	92.5	
N2a	6	2.5	6	3.3			
N2b	77	31.0	26	14.4	51	76.1	
N2c	21	8.5	10	5.5	11	16.4	
Extranodal extension							
No positive node	103	41.6	102	56.4	1	1.5	**<.001**
Positive	37	14.9	13	7.2	24	35.8	
Negative	108	43.5	66	36.4	42	62.7	
LNN							
Mean ± SD	32.02 ± 16.77		32.74 ± 16.78		30.12 ± 16.71		
Range	1 ~ 100		1 ~ 95		8 ~ 100		
pLNN							
Mean ± SD	1.90 ± 3.20		0.78 ± 1.16		4.94 ± 4.68		
Range	0 ~ 35		0 ~ 7		1 ~ 35		
PNI							
Positive	33	13.3	25	13.8	8	11.9	.685
Negative	215	86.7	156	86.2	59	88.1	
Pathological margins							
Positive	0	0	0	0	0	0	
Negative	248	100	181	100	67	100	
pTNM							
0	11	4.4	11	6.1			**<.001**
I	12	4.8	12	6.6			
II	21	8.5	21	11.6			
III	75	30.2	69	38.1	6	9.0	
IV	129	52.0	68	37.6	61	91.0	

Abbreviations: ENE, extranodal extension; LNN, lymph nodes numbers; LNR, lymph node ratio; pLNN, number of positive lymph nodes excised; PNI, perineural invasion.

*P value from Chi-square test was reported to compare baseline characteristics between the low-risk and high-risk LNR groups.

Bold values indicate statistical values.

2.4 | TCGA data collection

A public retrospective data set specified for OSCC was collected by The Cancer Genome Atlas (TCGA) from cBioPortal (http://www.cbioportal.org/). Raw data (Supplementary excel file S1) were manually checked and integrated. A diagram showing patient selection is provided in Figure S1. Among the 343 OSCC patients who underwent primary lesion resection and neck dissection from 1994-2013,

a total of 91 OSCC patients with positive lymph node metastases were enrolled in this study. The patient demographic and clinical data are presented in Table S1.

2.5 | Statistical analysis

Continuous variables are represented as the mean ± SD. Chi-square or Pearson's chi-square test was used to analyze the clinical and histologic data. A receiver operating characteristic (ROC) curve with area under the curve (AUC) was used to calculate the best cutoff value for the LNR. Univariate and multivariate Cox regression models were used to investigate the hazard ratio (HR). Effect modifications were conducted by univariate Cox regression analysis when the HR was significant ($P < .05$). The variables selected into the multivariate model were determined by univariate Cox regression analysis and clinical evaluation. Collinearity diagnostics were used to exclude the multicollinearity among variables (Tolerance > 0.1 and Variance Inflation Factor (VIF) < 10 mean no multicollinearity among covariates). All variables ($P < .10$, tolerance > 0.1 and VIF < 10) were included in the multivariate Cox regression model (forward method) to further evaluate better prognostic predictors. The Kaplan-Meier method and log-rank test were used for survival analysis. All hypothesis-generating tests were two-sided at a significance level of 0.05. Data were analyzed with IBM SPSS Statistics 23 (IBM Corporation, Armonk, NY, USA).

3 | RESULTS

3.1 | Patients

Among the 248 OSCC patients, the total number of lymph nodes excised and examined was 7941, and the median number of lymph nodes excised was 29 with a range of 1-100. There were 472 (5.9%) lymph nodes that were pathologically confirmed to have positive metastases. One-hundred and forty-five (58.4%) patients (79 in the control group and 66 in the experimental group) were pathologically confirmed to have lymph node metastases (pN+). Table 1 and Table S2 present the pathological and clinical data of these patients.

The median follow-up time was 80 months (range, 3.2-93 months). During the follow-up period, death, tumor recurrence, and tumor distant metastasis were recorded in 109 (44%), 125 (50.4%), and 109 patients (44%), respectively. The 5-year OS, DFS, DSS, LRFS, and DMFS were 58.8%, 51.5%, 63.0%, 53.1%, and 58.4%, respectively.

3.2 | Lymph node ratio predicts clinical outcomes in patients with locally advanced OSCC

The postoperative pathological examinations showed that 103 patients (41.5%) were at pN0 stage, 41 (16.5%) at pN1 stage, and 104 (42.0%) at pN2 stage. Among the 145 pN + patients, there were 35.1 ± 17.7 (range, 8-100) lymph nodes isolated and pathologically examined per patient; 3.2 ± 3.6 (range, 1-35) lymph nodes were metastatic. For the continuous variable of LNR, the median value of LNR was 0.070 with a range from 0 to 0.7609; the cutoff value of 0.076 for predicting DFS was calculated on the basis of ROC curves ($P < .001$, and AUC = 0.628, Figure 1). The same cutoff value was confirmed in OS, DSS, LRFS, and DMFS. Subsequently, all patients were allocated into the high LNR (>7.6%, n = 67), low LNR (≤ 7.6%, n = 78), or LNR = 0 (n = 103) groups.

Survival analysis between the patients with pN + and pN0 showed that the patients with pN0 had significantly better OS, DFS, DSS, LRFS, and DMFS than patients with pN+ (Figure S2). The 5-year OS, DFS, DSS, LRFS and DMFS were 68.9%, 62.1%, 76.7%, 64.1%, and 68.0% in the patients with pN0, and 51.6%, 43.9%, 55.5%, 45.3%, and 51.6% in the patients with pN+, respectively.

Then survival analysis indicated that both the patients with LNR = 0 and LNR ≤ 7.6% had significantly better OS, DFS, DSS, LRFS, and DMFS than patients with LNR > 7.6%. The difference in OS, DFS, DSS, LRFS, and DMFS between the patients with LNR = 0 and LNR ≤ 7.6% was not significant (OS, $P = .190$; DFS, $P = .088$; DSS, $P = .055$; LRFS, $P = .104$; DMSF, $P = .183$; Figure S3). Therefore, the patients with a LNR = 0 and LNR ≤ 7.6% were combined as the low-risk LNR subset, while patients with LNR > 7.6% were considered to be the high-risk LNR subset. The patients with low-risk LNR (n = 181) had significant better clinical outcomes with respect to OS (HR = 2.321, 95%CI:1.577-3.414, $P < .001$), DFS (HR = 1.996, 95%CI:1.386-2.874, $P < .001$), DSS (HR = 2.443, 95%CI:1.611-3.705, $P < .001$), LRFS (HR = 2.021, 95%CI:1.397-2.924, $P < .001$), and DMFS (HR = 2.280, 95%CI:1.550-3.353, $P < .001$) compared to patients with high-risk LNR (n = 67; Figure 2).

According to the chi-square test, the patients with high-risk LNR were associated with increased pN stage ($P < .001$) and positive ENE ($P < .001$; Table 1).

3.3 | Lymph node ratio as independent predictor for survival in pN + patients

In order to evaluate the predictive factor for prognosis considering both the number of positive lymph nodes and total number of lymph nodes, 145 pN + patients were analyzed while the pN0 patients were not included in the following analysis. Among the 145 pN + patients, 79 patients in the control group were firstly analyzed using the univariate and multivariate Cox model analyses; then the 145 patients in both the control and experimental groups were analyzed for verification.

In the 79 patients in the control group, the clinical and pathological covariates were all evaluated using univariate Cox model analysis: pTNM stage (stage IV vs stage III, HR = 4.495, 95%CI:1.387-14.565, $P = .012$) and LNR (high-risk vs low-risk, HR = 2.110, 95%CI:1.159-3.84, $P = .015$) were significant predictors for OS (Table 2); pTNM stage (stage IV vs stage III, HR = 2.142, 95%CI:1.136-4.041, $P = .019$) and LNR (high-risk vs low-risk, HR = 1.899, 95%CI:1.078-3.346, $P = .026$) for DFS; pTNM stage (stage IV vs stage III, HR = 3.900, 95%CI:1.196-12.714, $P = .024$) and LNR (high-risk vs low-risk,

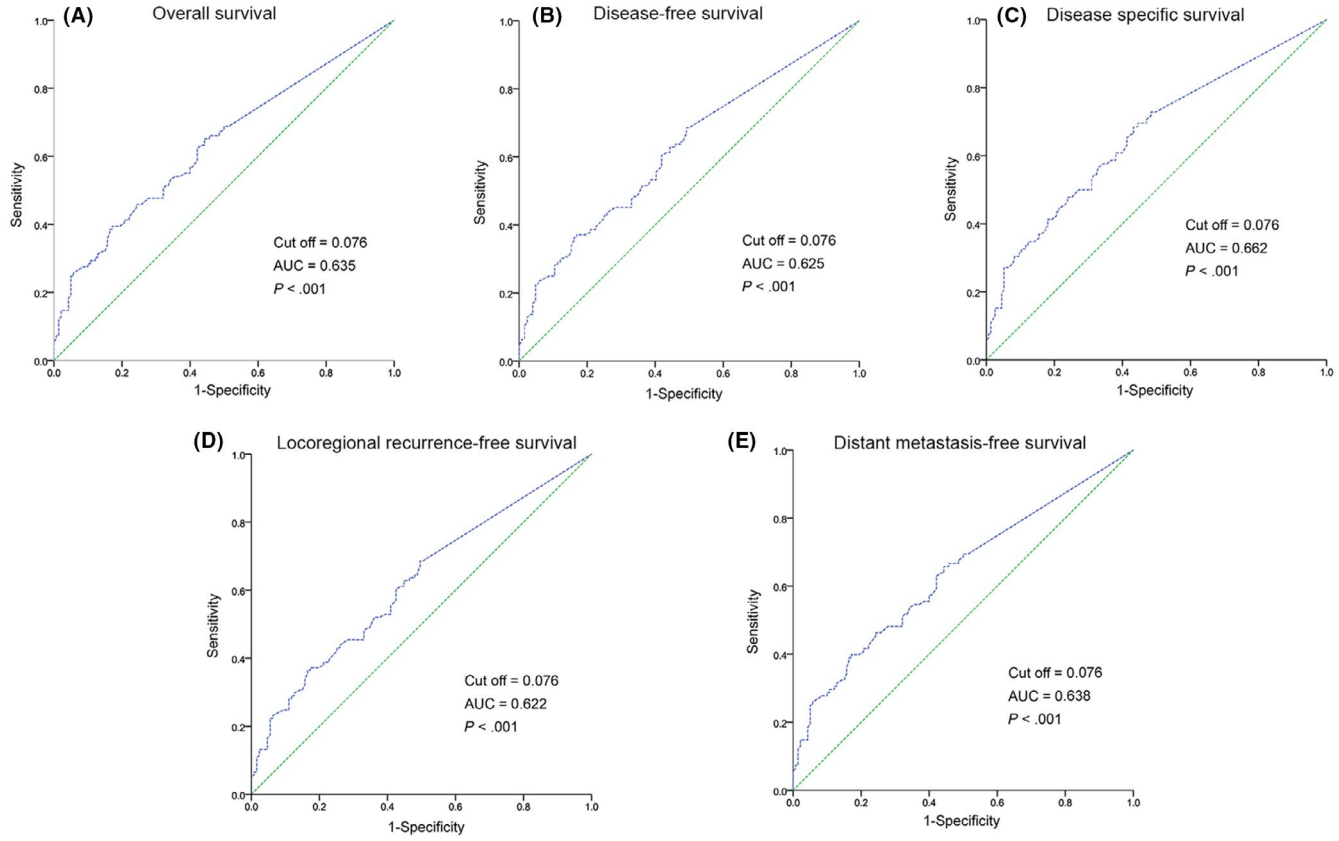

FIGURE 1 Receiver operating characteristic curve for lymph node ratio analysis using a cutoff value of 0.0076 according to disease-free survival (B), and confirmed in overall survival (A), disease-specific survival (C), locoregional recurrence-free survival (D), and distant metastasis-free survival (E). AUC, area under the curve

HR = 2.019, 95%CI:1.072-3.805, *P* = .030) for DSS; pTNM stage (stage IV vs stage III, HR = 2.698, 95%CI:1.065-6.833, *P* = .036) and LNR (high-risk vs low-risk, HR = 1.913, 95%CI:1.073-3.408, *P* = .028) for LRFS; pTNM stage (stage IV vs stage III, HR = 4.153, 95%CI:1.281-13.464, *P* = .018) and LNR (high-risk vs low-risk, HR = 2.056, 95%CI:1.129-3.744, *P* = .018) for DMFS.

Significant univariates and ENE were included in the multivariate Cox model analysis, and only LNR (high-risk vs low-risk) was an independent predictive factor for OS (HR = 2.038, 95%CI: 1.012-4.102, *P* = .046) and DMFS (HR = 2.099, 95%CI: 1.321-3.337, *P* = .049) (Table 3).

Then, the 145 pN + patients (including those in the experimental group) were analyzed using univariate and multivariate Cox models. Univariate Cox model analysis showed that LNR (high-risk vs low-risk) was a significant predictor for OS (*P* = .006), DFS (*P* = .030), DSS (*P* = .013), LRFS (*P* = .027), and DMFS (*P* = .007); pT (T3 and T4 vs T1), pTNM, and ENE were significant predictors for OS, DFS, DSS, LRFS, and DMFS (Table S3). All covariates (*P* < .10 and tolerance > 0.1) were included in the multivariate Cox model (forward method) analysis to further evaluate better prognostic predictors, LNR (high-risk vs low-risk, HR = 2.153, 95%CI: 1.354-3.423, *P* = .001; HR = 1.736, 95%CI: 1.129-2.669, *P* = .012; HR = 2.056, 95%CI: 1. 262-3.350, *P* = .004; HR = 1.763, 95%CI:1.141-2.725, *P* = .011; HR = 2.099, 95%CI:1.321-3.337, *P* = .002) and pT stage (*P* < .001) were independent predictive factors for OS, DFS, DSS, LRFS, and DMFS (Table S4).

3.4 | Validation of our cutoff value of LNR in the TCGA database

To perform external validation of our LNR cutoff value, 91 OSCC patients with pN + from the TCGA database were enrolled. Among the 91 OSCC patients, there were 41 ± 20.6 (range, 9-101) lymph nodes excised and examined, and 2 ± 2.7 (range, 1-14) lymph nodes had positive metastases. The mean LNR was 0.0779 (range, 0.013-0.500); Table S1 presents the demographic and clinical data of these patients. The 91 patients with pN + were subsequently allocated into the following two categories: LNR > 7.6% (n = 30); and LNR ≤ 7.6% (n = 61). Using the Kaplan-Meier method and univariate analyses, LNR (cutoff value = 0.076) was a predictive factor for OS (HR = 3.823, 95%CI:1.839-7.948, *P* < .001) and DFS (HR = 3.172, 95%CI:1.713-5.872, *P* < .001), respectively (Figure S4 and Table S5).

4 | DISCUSSION

Based on our results, the OSCC patients with high-risk LNR had a worse clinical outcome with respect to OS, DFS, DSS, LRFS, and DMFS than patients with low-risk LNR. Multivariate analysis showed that high-risk LNR was an independent worse indicator for OS, DFS, DSS, LRFS, and DMFS among the pN + patients, which was more

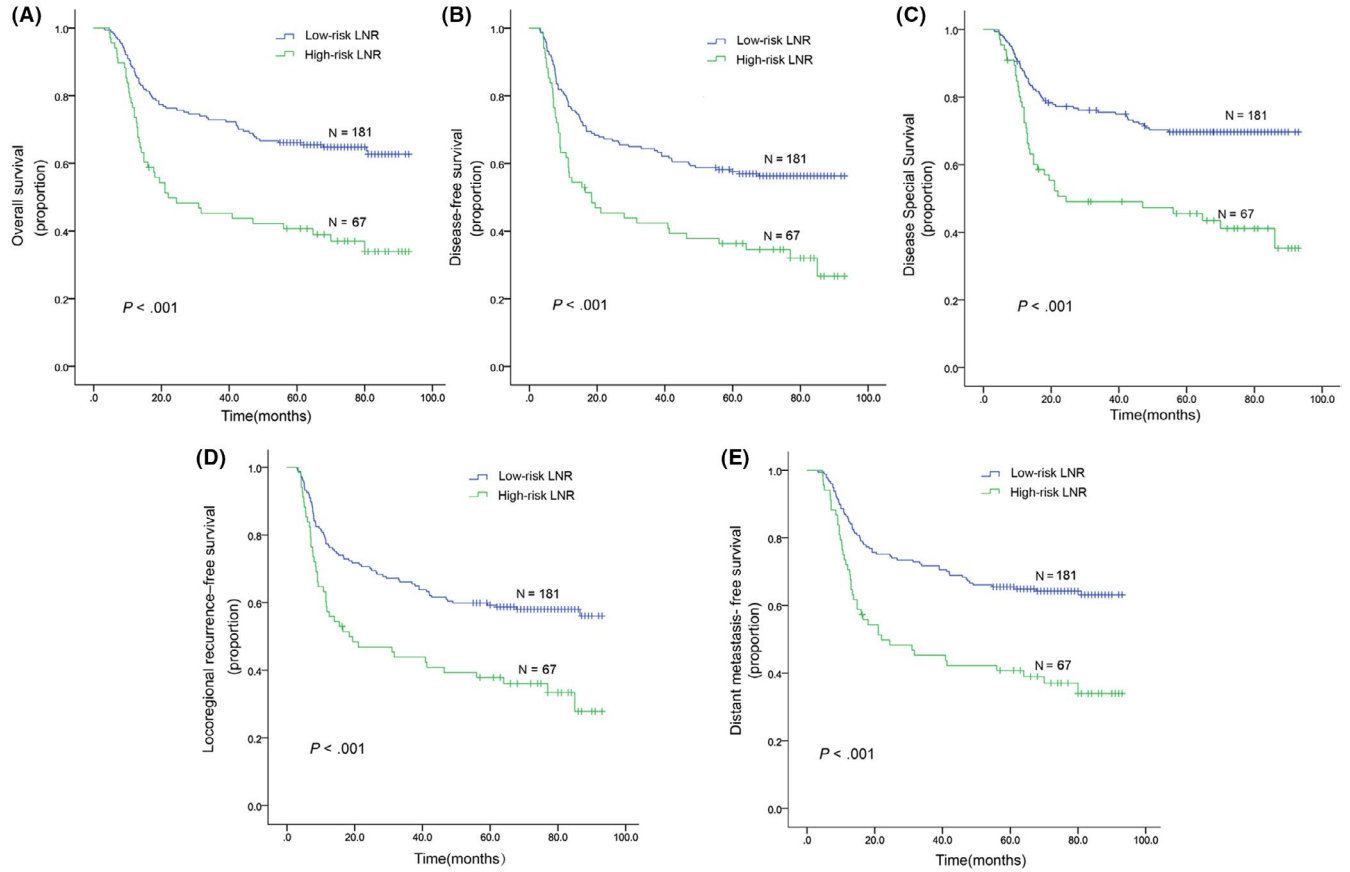

FIGURE 2 Survival analysis between patients with a low- and high-risk LNR. A, Overall survival, (B) disease-free survival, (C) disease-specific survival, (D) locoregional recurrence-free survival, (E) distant metastasis-free survival. LNR, lymph node ratio

accurate than pN stage. The cutoff value of LNR of 7.6% was also verified with similar results using an open TCGA database, high-risk LNR indicating worse OS and DFS.

Lymph node status plays a vital role in predicting survival outcomes of OSCC patients. Indeed, patients with positive lymph node metastases have a poor prognosis and need postoperative adjuvant therapy; however, to stratify the pN + patients to receive more treatment is not enough.[6,16] There is an ongoing debate regarding the predictive value of the conventional pN stage and evaluating the number, size, and distribution of positive lymph nodes.[17,18] To improve the accuracy of prognostic evaluation, ENE status has been added in the newly published 8th edition of the AJCC Cancer Staging Manual, in which the pN stage has been modified by inclusion of ENE status. Despite this, given that the technical performance on identifying metastasis of lymph nodes may vary among surgeons [19,20] and pathologists,[21,22] LNR has emerged as a superior prognostic factor for OSCC as well as other solid tumors.[5-9,23] Three factors of LNR have been taken into account in the conventional pN stage, including (a) the true number of positive lymph nodes, (b) the actual number of lymph nodes excised, and (c) the completeness of the pathological analysis.[4] In the present study, our analysis demonstrates that LNR is an independent prognostic factor in patients with locally advanced OSCC, which is consistent with the previous studies that LNR is better than the conventional pN stage for prognostic evaluation in OSCC.[4,6,24] On the aspect of pT stage, when we analyzed the pN + patients, who did not receiving TPF induction chemotherapy, pT stage was not a significant predictor for clinical outcomes; but when we included the patients received TPF induction chemotherapy, the patients at advanced pT (pT3/pT4) stage had worse prognosis compared with those at pT1 stage. This might be due to the fact that some patients with a good pathological response to TPF induction chemotherapy have a better prognosis. To verify the cutoff value of LNR in this study, an open TCGA database was used and the results indicated that our cutoff value of LNR was trustworthy.

In our study, although we found that the prognostic difference was not significant between the patients with a LNR = 0 and a LNR ≤ 7.6%, it should be further confirmed by clinical trials to determine whether or not postoperative adjuvant treatment should be the same between these patients. For patients with a high-risk LNR, more aggressive treatment might improve the prognosis[23]; however, in patients with OSCC, it is still unknown whether or not patients with a high-risk LNR need more aggressive treatment.[6] Our previous study showed that the cN2 OSCC patients could benefit from TPF induction chemotherapy.[10,11] In this retrospective study, pathological LNR might identify the potential population who could benefit from TPF induction chemotherapy. Despite this, it might be better to

TABLE 2 Univariate analysis of prognostic factors in the 79 pN + patients in the control group

	Overall survival			Disease-free survival			Disease-specific survival			Locoregional recurrence-free survival			Distant metastasis-free survival		
Variables	**HR**	**95%CI**	***P***	**HR**	**95%CI**	***P***	**HR**	**95%CI**	***P***	**HR**	**95%CI**	***P***	**HR**	**95%CI**	***P***
Sex	1.296	0.677-2.483	.434	1.009	0.555-1.833	.977	1.207	0.611-2.385	.588	1.031	0.558-1.905	.921	1.324	0.692-2.536	.397
Age	1.281	0.683-2.403	.441	1.278	0.706-2.316	.418	1.103	0.556-2.188	.778	1.236	0.673-2.272	.495	1.261	0.673-2.365	.469
pT stage			.123			.214			.130			.350			.137
T1		Ref			Ref			Ref			Ref			Ref	
T2	1.863	0.420-8.256	.413	1.374	0.398-4.749	.615	1.608	0.356-7.258	.537	1.367	0.396-4.725	.621	1.843	0.330-8.168	.421
T3	3.046	0.706-13.143	.135	2.339	0.695-7.880	.170	2.547	0.581-11.166	.215	1.936	0.570-6.576	.290	3.076	0.407-13.269	.132
T4	3.761	0.831-17.062	.086	2.429	0.676-8.723	.174	3.711	0.817-16.848	.089	2.423	0.675-8.701	.175	3.705	0.818-16.774	.089
pN stage			.062			.265			.113			.353			.084
N1	Ref	Ref		Ref			Ref								
N2a	0.329	0.044-2.476	.977	1.189	0.263-5.369	.822	—	—	.978	1.245	0.276-5.620	.776	—	—	.976
N2b	2.084	1.001-4.337	.050	1.953	0.972-3.924	.060	1.900	0.876-4.120	.104	1.813	0.897-3.666	.097	1.980	0.951-4.124	.068
N2c	2.135	0.803-5.677	.129	1.854	0.717-4.797	.203	2.197	0.813-5.936	.121	1.925	0.744-4.982	.177	2.019	0.758-4.374	.160
Smoke status	1.582	0.874-2.862	.130	1.462	0.832-2.561	.185	1.536	0.819-2.882	.181	1.408	0.794-2.496	.241	1.629	0.900-2.947	.107
Drink status	1.386	0.760-2.529	.287	1.416	0.801-2.506	.232	1.284	0.673-2.447	.448	1.322	0.738-2.369	.348	1.435	0.786-2.618	.239
Site			.617			.749			.358			.841			.695
PNI (Yes vs No[a])	1.125	0.555-2.282	.743	1.117	0.570-2.188	.747	1.323	0.644-2.717	.446	1.038	0.515-2.092	.916	1.087	0.536-2.202	.818
ENE (Yes vs No[a])	1.459	0.780-2.727	.237	1.485	0.823-2.680	.189	1.515	0.786-2.919	.215	1.653	0.910-3.005	.099	1.391	0.743-2.601	.302
pTNM stage (IV vs III)	4.495	1.387-14.565	**.012**	2.142	1.136-4.041	**.019**	3.900	1.196-12.714	**.024**	2.698	1.065-6.833	**.036**	4.153	1.281-13.464	**.018**
LNR (high- vs low-risk[b])	2.110	1.159-3.841	**.015**	1.899	1.078-3.346	**.026**	2.019	1.072-3.805	**.030**	1.913	1.073-3.408	**.028**	2.056	1.129-3.744	**.018**

Abbreviations: ENE, extranodal extension; LNR, lymph node ratio; PNI, perineural invasion; Ref, reference category.

[a]Yes: positive, No: negative;

[b]high-risk LNR: LNR > 7.6%, low-risk LNR: LNR ≤ 7.6%.

Bold values indicate statistical values.

TABLE 3 Multivariate analysis of prognostic factors in the 79 pN + OSCC patients in the control group

Variables	Overall survival			Disease-free survival			Disease-specific survival			Locoregional recurrence-free survival			Distant metastasis-free survival		
	HR	95%CI	*P*	HR	95%CI	*P*	HR	95%CI	*P*	HR	95%CI	*P*	HR	95%CI	*P*
pT stage	–	–	.139			.215			.107	–	–	.270			.133
pN stage	–	–	.669	–	–	.657			.752	–	–	.528	–	–	.703
pTNM	–	–	.117	–	–	.266			.224	–	–	.249	–	–	.156
ENE	–	–	.447			.361			.441			.192			.555
LNR	2.038	1.012-4.102	**.046**	1.736	1.129-2.669	**.060**			.076	1.763	1.141-2.725	**.053**	2.099	1.321-3.337	**.049**

Abbreviations: ENE, extranodal extension; LNR, lymph node ratio.
Bold values indicate statistical values.

have the pretreatment clinical LNR records for prognostic analysis. Further investigations are warranted to confirm this result.

Recent studies have shown that ENE status has been reported as a major prognostic factor.[25-27] In this study, we also found that the ENE status was a prognostic factor in pN + OSCC patients. However, the significance of the extent of ENE is controversial, particularly when patients receive postoperative adjuvant therapy.[15] The results of multivariate analysis of ENE and LNR are contradictory. Adding LNR into the model analysis resulted in a loss of independent prognostic ability of ENE[28,29]; while, both ENE and LNR have also been reported to have prognostic significance based on multivariate studies.[6,30] Our multivariate analysis results showed that LNR might be a superior prognostic predictor compared with ENE status in pN + OSCC patients.

There are still some limitations to our study. Firstly, the dataset is retrospective, TPF induction chemotherapy before surgery may change the status of lymph node metastasis. Secondly, the exact total number of lymph nodes from imaging examinations might be inaccurate in this study. On the decision of TPF induction chemotherapy depending on LNR, we suggest to evaluate the pretreatment clinical LNR by pretreatment imaging examinations in the future investigations.

5 | CONCLUSIONS

Oral squamous cell carcinoma patients with a high-risk LNR have worse clinical outcomes than patients with a low-risk LNR, and high-risk LNR is an independent negative factor for clinical outcomes.

ACKNOWLEDGEMENTS

This study was supported by National Natural Science Foundation of China (81672660), Shanghai Municipal Commission of Health and Family Planning (2018BR41), Clinical Research Project of Multi-Disciplinary Team, Ninth People's Hospital, Shanghai Jiao Tong University School of Medicine (201916), Two-hundred Talent, Shanghai Jiao Tong University School of Medicine (20191915).

CONFLICT OF INTEREST

The authors declare no conflict of interest regarding the publication of this paper.

AUTHOR CONTRIBUTIONS

Tong-Chao Zhao: Formal analysis, Investigation, Methodology, Visualization, Writing–original draft, Writing–review and editing. **Si-Yuan Liang**: Formal analysis, Investigation, Methodology, Validation, Visualization, Writing–original draft, Writing–review and editing. **Wu-Tong Ju**: Conceptualization, Data curation, Project administration, Resources, Software, Writing–original draft, Writing–review and editing. **Yong Fu**: Data curation, Methodology, Writing–original draft, Writing–review and editing. **Zhi-Hang Zhou**: Methodology, Writing–original draft, Writing–review and editing. **Li-Zhen Wang**: Data curation, Investigation, Methodology, Resources, Writing–original draft, Writing–review and editing. **Jiang Li**: Conceptualization, Project administration, Resources, Supervision, Writing–original

draft, Writing–review and editing. **Chen-Ping Zhang**: Project administration, Resources, Writing–original draft, Writing-review and editing. **Zhi-Yuan Zhang**: Resources, Supervision, Writing–original draft, Writing–review and editing. **Lai-Ping Zhong**: Conceptualization, Data curation, Funding acquisition, Project administration, Resources, Supervision, Writing–original draft, Writing–review and editing.

ORCID

Lai-Ping Zhong https://orcid.org/0000-0002-5528-9169

REFERENCES

1. Bando E, Yonemura Y, Taniguchi K, Fushida S, Fujimura T, Miwa K. Outcome of ratio of lymph node metastasis in gastric carcinoma. *Ann Surg Oncol*. 2002;9(8):775-784.
2. Berger AC, Sigurdson ER, LeVoyer T, et al. Colon cancer survival is associated with decreasing ratio of metastatic to examined lymph nodes. *J Clin Oncol*. 2005;23(34):8706-8712.
3. Woodward WA, Vinh-Hung V, Ueno NT, et al. Prognostic value of nodal ratios in node-positive breast cancer. *J Clin Oncol*. 2006;24(18):2910-2916.
4. Patel SG, Amit M, Yen TC, et al. Lymph node density in oral cavity cancer: results of the International Consortium for Outcomes Research. *Br J Cancer*. 2013;109(8):2087-2095.
5. Chow TL, Kwan W, Fung SC, Ho LI. Prognostic value of lymph node density in buccal squamous cell carcinoma. *Am J Otolaryngol*. 2017;38(5):529-532.
6. Feng Z, Xu QS, Wang C, et al. Lymph node ratio is associated with adverse clinicopathological features and is a crucial nodal parameter for oral and oropharyngeal cancer. *Sci Rep*. 2017;7(1):6708.
7. Lieng H, Gebski VJ, Morgan GJ, Veness MJ. Important prognostic significance of lymph node density in patients with node positive oral tongue cancer. *Anz J Surg*. 2016;86(9):681-686.
8. Reinisch S, Kruse A, Bredell M, Lubbers HT, Gander T, Lanzer M. Is lymph-node ratio a superior predictor than lymph node status for recurrence-free and overall survival in patients with head and neck squamous cell carcinoma? *Ann Surg Oncol*. 2014;21(6):1912-1918.
9. Safi AF, Grandoch A, Nickenig HJ, Zoller JE, Kreppel M. The importance of lymph node ratio for locoregional recurrence of squamous cell carcinoma of the tongue. *J Craniomaxillofac Surg*. 2017;45(7):1058-1061.
10. Zhong LP, Zhang CP, Ren GX, et al. Randomized phase III trial of induction chemotherapy with docetaxel, cisplatin, and fluorouracil followed by surgery versus up-front surgery in locally advanced resectable oral squamous cell carcinoma. *J Clin Oncol*. 2013;31(6):744-751.
11. Zhong LP, Zhang CP, Ren GX, et al. Long-term results of a randomized phase III trial of TPF induction chemotherapy followed by surgery and radiation in locally advanced oral squamous cell carcinoma. *Oncotarget*. 2015;6(21):18707-18714.
12. Woolgar JA. Detailed topography of cervical lymph-note metastases from oral squamous cell carcinoma. *Int J Oral Maxillofac Surg*. 1997;26(1):3-9.
13. Woolgar JA. The topography of cervical lymph node metastases revisited: the histological findings in 526 sides of neck dissection from 439 previously untreated patients. *Int J Oral Maxillofac Surg*. 2007;36(3):219-225.
14. Carter RL, Barr LC, O'Brien CJ, Soo KC, Shaw HJ. Transcapsular spread of metastatic squamous cell carcinoma from cervical lymph nodes. *Am J Surg*. 1985;150(4):495-499.
15. Woolgar JA, Rogers SN, Lowe D, Brown JS, Vaughan ED. Cervical lymph node metastasis in oral cancer: the importance of even microscopic extracapsular spread. *Oral Oncol*. 2003;39(2):130-137.
16. Goldstein DP, Bachar GY, Lea J, et al. Outcomes of squamous cell cancer of the oral tongue managed at the Princess Margaret Hospital. *Head Neck*. 2013;35(5):632-641.
17. Ryu IS, Roh JL, Cho KJ, Choi SH, Nam SY, Kim SY. Lymph node density as an independent predictor of cancer-specific mortality in patients with lymph node-positive laryngeal squamous cell carcinoma after laryngectomy. *Head Neck*. 2015;37(9):1319-1325.
18. Vinh-Hung V, Verkooijen HM, Fioretta G, et al. Lymph node ratio as an alternative to pN staging in node-positive breast cancer. *J Clin Oncol*. 2009;27(7):1062-1068.
19. Henick DH, Silver CE, Heller KS, Shaha AR, El GH, Wolk DP. Supraomohyoid neck dissection as a staging procedure for squamous cell carcinomas of the oral cavity and oropharynx. *Head Neck*. 1995;17(2):119-123.
20. Agrama MT, Reiter D, Cunnane MF, Topham A, Keane WM. Nodal yield in neck dissection and the likelihood of metastases. *Otolaryngol Head Neck Surg*. 2003;128(2):185-190.
21. van den Brekel MW, van der Waal I, Meijer CJ, Freeman JL, Castelijns JA, Snow GB. The incidence of micrometastases in neck dissection specimens obtained from elective neck dissections. *Laryngoscope*. 1996;106(8):987-991.
22. Kim Y, Park SH, Kim KM, et al. The influence of metastatic lymph node ratio on the treatment outcomes in the adjuvant chemoradiotherapy in stomach tumors (ARTIST) trial: a phase III trial. *J Gastric Cancer*. 2016;16(2):105-110.
23. Safi AF, Kauke M, Grandoch A, et al. The importance of log odds of positive lymph nodes for locoregional recurrence in oral squamous cell carcinoma. *Oral Oncol*. 2017;72(1):48-55.
24. Maxwell JH, Ferris RL, Gooding W, et al. Extracapsular spread in head and neck carcinoma: impact of site and human papillomavirus status. *Cancer*. 2013;119(18):3302-3308.
25. Mermod M, Tolstonog G, Simon C, Monnier Y. Extracapsular spread in head and neck squamous cell carcinoma: a systematic review and meta-analysis. *Oral Oncol*. 2016;62(1):60-71.
26. Suton P, Salaric I, Granic M, Mueller D, Luksic I. Prognostic significance of extracapsular spread of lymph node metastasis from oral squamous cell carcinoma in the clinically negative neck. *Int J Oral Maxillofac Surg*. 2017;46(6):669-675.
27. Ebrahimi A, Clark JR, Zhang WJ, et al. Lymph node ratio as an independent prognostic factor in oral squamous cell carcinoma. *Head Neck*. 2011;33(9):1245-1251.
28. Chen CC, Lin JC, Chen KW. Lymph node ratio as a prognostic factor in head and neck cancer patients. *Radiat Oncol*. 2015;10:181.
29. Majercakova K, Valero C, Lopez M, et al. Postoperative staging of the neck dissection using extracapsular spread and lymph node ratio as prognostic factors in HPV-negative head and neck squamous cell carcinoma patients. *Oral Oncol*. 2018;77(1):37-42.
30. Amar A, Rapoport A, Curioni OA, Dedivitis RA, Cernea CR, Brandao LG. The density of metastatic lymph node as prognostic factor in squamous cell carcinoma of the tongue and floor of the mouth. *Braz J Otorhinolaryngol*. 2012;78(3):86-90.

SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section.

How to cite this article: Zhao T-C, Liang S-Y, Ju W-T, et al. High-risk lymph node ratio predicts worse prognosis in patients with locally advanced oral cancer. *J Oral Pathol Med*. 2020;49:787–795. https://doi.org/10.1111/jop.13043