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Original Article

The status of perineural invasion predicts the outcomes of postoperative radiotherapy in locally advanced esophageal squamous cell carcinoma

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Abstract: Background: Prognosis of locally advanced esophageal squamous cell carcinoma (ESCC) remains dismal even after curative resection and adjuvant radiotherapy. New biomarkers for predicting prognosis and treatment outcomes are needed for improved treatment stratification of patients with locally advanced ESCC. The prognostic and treatment predictive significance of perineural invasion (PNI) in the locally advanced ESCC remains unclear. This study aimed to examine the effect of PNI on the outcomes of locally advanced ESCC patients after curative resection with or without postoperative radiotherapy (PORT). Patients and methods: We retrospectively reviewed 262 consecutive locally advanced ESCC patients who underwent curative resection. Tumors sections were re-evaluated for PNI by an independent pathologist blinded to the patients' outcomes. Overall survival (OS) and disease-free survival (DFS) were determined using the Kaplan-Meier method; univariate log-rank test and multivariate Cox proportional hazard model were used to evaluate the prognostic value of PNI. Results: Finally, 243 patients were analyzed and enrolled into this study, of which 132 received PORT. PNI was identified in 22.2% (54/243) of the pathologic sections. The 5-year DFS was favorable for PNI-negative patients versus PNI-positive patients (21.3% *vs.* 36.7%, respectively; $P = 0.005$). The 5-year OS was 40.3% for PNI-negative patients versus 21.7% for PNI-positive patients ($P < 0.001$). On multivariate analysis, PNI was an independent prognostic factor. In a subset analysis for patients received PORT, PNI was evaluated as a prognostic predictor as well ($P < 0.05$). In contrast to patients without PORT, PORT couldn't improve the disease recurrence and survival in locally advanced ESCC patients with PNI-positive ($P > 0.05$). Conclusions: PNI could serve as an independent prognostic factor and prognosticate treatment outcomes in locally advanced ESCC patients. The PNI status should be considered when stratifying high-risk locally advanced ESCC patients for adjuvant radiotherapy. Future prospective study is warranted to confirm our results.

Keywords: Biomarkers, perineural invasion, prognosis, esophageal squamous cell carcinoma, postoperative radiotherapy

Introduction

Great progress has been made for the treatment of esophageal cancer, whereas clinical outcome of locally advanced esophageal squamous cell carcinoma (ESCC) remains disappointed with a 5-year survival rate not exceeding 30% in China [1]. Surgery is the treatment of choice for resectable ESCC. However the outcome of surgery alone for tumors invading beyond the muscularis propria or involving locoregional lymph nodes is poor. As of now, the benefit of adjuvant radiotherapy remains controversial for ESCC patients after curative resection. Presently no definite conclusions can be drawn on the basis of the available evidence [2]. Therefore, it remains difficult to determine the populations who would benefit from adjuvant radiotherapy in high-risk locally advanced ESCC patients. Therefore, it is crucially important to identify the high-risk factors closely associated with disease recurrence and poor prognosis, which can serve as the basis of selection of locally advanced ESCC patients for adjuvant therapy.

It is well known that tumor cells interact with the non-malignant cells and stromal elements

that constitute the tumor microenvironment [3]. Peripheral nerves surrounding or within the tumor cells, far from being mere bystanders, have been recognized as potentially important components of the tumor microenvironment [4]. However less emphasis has been laid on the significance of perineural invasion (PNI) in tumor dissemination and metastasis [5]. Regarded as a local spread route of tumor, PNI has been reported to be significantly associated with poor prognosis in several cancers [6-8]. Previous studies have also evaluated the prognostic significance of PNI in ESCC, and no significant prognostic value is found [9-11]. However, it has been reported recently that the PNI status corresponded to the tumor progression and may function as an independent prognostic indicator [12]. Therefore, this uncertainty of prognostic significance of PNI in ESCC patients is worth further clarification. In this large cohort of ESCC patients, we aimed to evaluate the prognostic significance of PNI status in locally advanced ESCC after curative resection, especially in patients received adjuvant radiotherapy. Moreover, the association of PNI with other clinic pathological factors and the effect of PNI on treatment failure after operative radiotherapy were also assessed.

Patients and methods

Patients

This study was approved by our Institutional Review Board, according to the Declaration of Helsinki. In this study locally advanced ESCC was defined as tumor invading beyond the muscularis propria or involving more than 2 locoregional lymph nodes (pT3-4 or N2-3) without distant metastasis at diagnosis. 262 consecutive patients with locally advanced ESCC who underwent radical esophagectomy (R_0) and mediastinal and abdominal lymph node dissection (conventional 2-field dissection) without preoperative radiotherapy and/or chemotherapy were retrospectively reviewed from January 2005 to December 2010. 14 patients were excluded because of incomplete resection and/or perioperative death and/or sections no longer available for analysis. And 5 patients were lost to follow-up. Finally, 243 patients were included into this study, of which 54.3% (132/243) of patients received PORT. Data about age, demographics, tumor location, staging, pathology, adjuvant radiotherapy and survival outcomes were obtained with the approval of our Institutional Review Board. The observation time in this study was the interval from surgical resection date to the last contact (death or last follow-up).

Radiotherapy

Three-dimensional conformal radiotherapy was performed with the dose of 50 to 54 Gy in 25 to 27 fractions for all patients four weeks after surgery. Irradiation target volume encompassed primary tumor bed, anastomosis and mediastinal and/or supraclavicular lymph node regions. Regarding the organs at risk, the maximum doses to the spinal cord was set as 45 Gy. In addition, the volume of lung received 20 Gy and 30 Gy was set no more than 28% and 20%, respectively. The mean dose to lung was set no more than 13 Gy. The volume of heart and thoracic stomach received 40 Gy was set no more than 50%.

Histopathologic evaluation

Hematoxylin-eosin stained sections were re-evaluated for the PNI status by an experienced pathologist who was blinded to any patient information, such as tumor stage and clinical outcome. At least four representative hematoxylin-eosin stained sections of each patient were studied in this cohort. PNI was defined as tumor cells within any layer of the nerve sheath or tumor close to the nerve and involving at least 33% of the nerve circumference [13].

Statistical analysis

All statistical analyses were performed using SPSS 13.0 software (SPSS, Inc., Chicago, IL). The relationship between PNI and clinicopathological factors was analyzed by the chi-square test. Overall and disease-free survival rates were calculated by the Kaplan-Meier method; univariate log-rank test and multivariate Cox proportional hazard model were used to evaluate the effect of PNI and other clinicopathological characteristics on disease-free survival (DFS) and overall survival (OS). *P*-value < 0.05 from the two-sided test was considered to be statistically significant.

Results

Patients and clinical characteristics

Patient characteristics and pathologic variables are shown in **Table 1**. 243 patients were

Table 1. Correlation of PNI with clinicopathological features in locally advanced ESCC patients

	N	PNI-positive	PNI-negative	*P*
Age				0.280
< 60	117	22 (18.8%)	95 (81.2%)	
≥ 60	126	32 (25.4%)	94 (74.6%)	
Sex				0.702
Male	194	42 (21.6%)	152 (78.4%)	
Female	49	12 (24.5%)	37 (75.5%)	
Location				0.475
Proximal esophagus	18	2 (11.1%)	16 (88.9%)	
Mid esophagus	165	39 (23.6%)	126 (76.4%)	
Distal esophagus	60	13 (21.7%)	47 (78.3%)	
Differentiation				0.365
Well	4	0 (0%)	4 (100%)	
Moderately	106	21 (19.8%)	85 (80.2%)	
Poorly	133	33 (24.8%)	100 (74.2%)	
Blood vessel invasion				0.758
Absent	127	27 (21.3%)	100 (78.7%)	
Present	116	27 (23.3%)	89 (76.7%)	
T stage				0.043
pT1-2	51	3 (9.6%)	48 (90.4%)	
pT3-4	192	51 (26.5%)	141 (73.5%)	
N stage				0.442
pN0	106	21 (19.8%)	85 (81.2%)	
pN^+	137	33 (24.1%)	104 (78.4%)	
AJCC stage				0.02
II	125	20 (16%)	105 (84%)	
III	118	34 (28.8%)	84 (71.2%)	
First failure location				
Local recurrence				0.021
Absent	124	20 (16.1%)	104 (83.9%)	
Present	119	34 (28.5%)	85 (71.5%)	
Distant metastasis				0.029
Absent	170	31 (18.2%)	139 (81.8%)	
Present	73	23 (31.5%)	50 (68.5%)	

eligible for this study: 194 males and 49 females with a median age at diagnosis of 60 years (range: 40-75 years). In accordance with the seventh American Joint Committee on Cancer (AJCC) Staging System 2010, the distribution of pathological stage was as follows: Stage II, 125 (51.4%); Stage III, 118 (48.6%).

PNI status in ESCC patients

The typical status of PNI in ESCC specimens are shown in **Figure 1**. The association between PNI status and clinicopathological features in the locally advanced ESCC patients is shown in **Table 1**. PNI was detected positive in 22.2% (54/243) of patients in this study, whereas of which only 10 (18.5%) patients were identified PNI-positive in the initial report. PNI positivity is closely correlated with known prognostic factors in ESCC. The depth of invasion (pT stage) and tumor stage were closely associated with the PNI positivity. In contrast, the relationship of PNI positivity with age, gender, tumor location, tumor differentiation, blood vessel invasion and pN stage was not detected.

The positivity of PNI was evidently increased with depth of tumor invasion and advanced stage. 9.6% of pT1-2 were PNI-positive compared with 26.5% of pT3-4 ($P = 0.043$). Stage III patients were near two times more likely to have PNI-positive tumors than stage II patients (odds ratio = 2.1; relative risk = 1.2; $P = 0.02$, **Table 1**). Furthermore, the status of PNI is also correlated with treatment failure including local recurrence and distant metastasis. 63% (34/54) of PNI-positive patients developed local relapse in comparison with 45% (85/189) of PNI-negative patients (odds ratio = 2.0; relative risk = 1.4; $P = 0.021$). Similarly 42% (23/54) of PNI-positive patients had distant metastasis compared to 25% (50/189) of PNI-negative patients (odds ratio = 2.1; relative risk = 1.6; $P = 0.029$).

PNI status predicts poor clinical outcomes in locally advanced ESCC patients

At a median follow-up of 2.4 years (range, 0.2 to 8.4 years), 158 patients (65%) died. At univariate analysis, PNI status, pT, pN, AJCC stage, grade, blood vessel invasion and PORT significantly influenced both DFS and OS (**Table 2**). Age, gender, and tumor location didn't significantly affect prognostic outcome. Overall, PNI-positive patients had a significantly increased rate of recurrence (5-year DFS rate, 21.3% vs. 36.7% for PNI-negative patients; $P = 0.005$; **Figure 2A**) and decreased OS (5-year survival rate, 21.7% vs. 40.3% for PNI-negative patients; $P < 0.001$; **Figure 2B**). The median DFS and OS

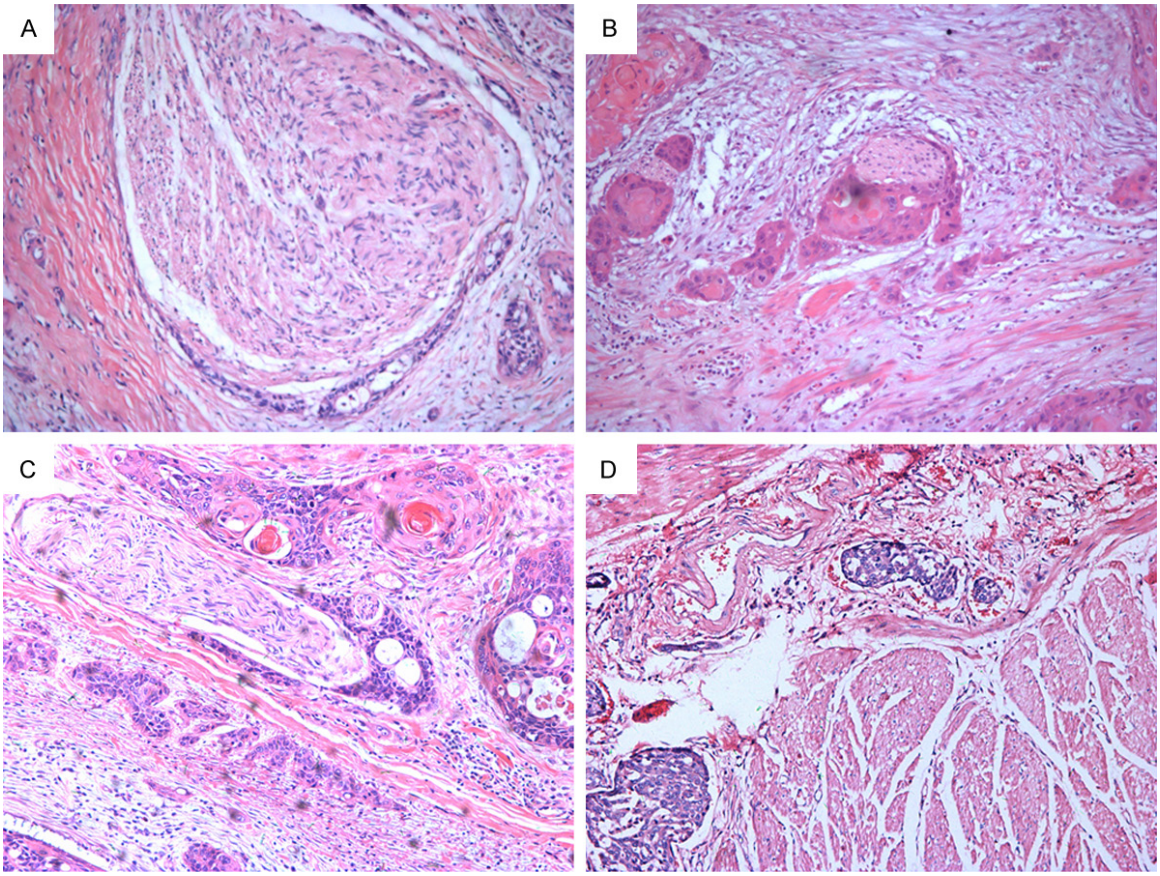

Figure 1. The typical status of perineural invasion (PNI) in ESCC specimens. Tumor cells located within perineural nerve sheath either (A) in epineurium or (B) in perineurium are evident example of PNI. (C) When tumor cells are not located inside of the nerve sheath but are in close proximity to the nerve, and involve at least 33% of the nerve circumference to diagnose PNI; (D) Without involvement of nerve means PNI-negative. (Hematoxylin-eosin staining, ×100).

time of PNI-positive patients were significantly worse than those of PNI-negative patients (12 and 15 months vs. 23 and 34 months, respectively; $P < 0.001$; **Figure 2A**, **2B**). Multivariate analysis using the Cox multiple regression model indicated that the status of PNI, PORT and AJCC stage were significant and independent prognostic factors for DFS and OS (**Table 3**).

Impact of PNI on survival of locally advanced ESCC patients received PORT

Among locally advanced ESCC patients received PORT, the 5-year DFS rate and the median DFS time were 46.3% and 43 months for PNI-negative patients compared with 24.8% and 17 months for PNI-positive patients ($P = 0.009$, **Figure 2C**). Moreover, PNI-positive locally advanced patients received PORT had an approximate DFS rate in contrast to locally advanced patients without PORT (24.3% vs. 23.2%, respectively; $P = 0.398$; **Figure 2C**). Similar results for OS were observed in locally advanced patients, the 5-year OS rate and the median OS time were significantly higher in PNI-negative patients than in PNI-positive patients (48% and 50 months vs. 24.8% and 24 months, respectively; $P = 0.005$; **Figure 2D**). Meanwhile, PNI-positive locally advanced patients received PORT had an approximate OS rate compared with locally advanced patients without PORT (24.3% vs. 27.7%, respectively; $P = 0.780$; **Figure 2D**).

Influence of PORT on survival of locally advanced ESCC patients with different PNI status

PORT didn't significantly improve the survival of locally advanced ESCC patients with PNI-positive. The 5-year DFS rate was 24.3% for patients received PORT compared with 18.5%

Table 2. DFS and OS in 243 locally advanced ESCC patients who underwent curative resection

	Patients	%	Disease-free Survival		Overall Survival	
			5 year (%)	*P*	5 year (%)	*P*
Age, years (mean, 60)				0.253		0.132
< 60	117	48.1	37.2		38.7	
≥ 60	126	51.9	29.8		33.9	
Sex				0.366		0.474
Male	194	79.8	31		34.5	
Female	49	20.2	42.9		42.9	
Location				0.776		0.862
Proximal esophagus	18	7.4	38.9		38.9	
Mid esophagus	165	67.9	31.3		34.9	
Distal esophagus	60	24.7	37.6		38.8	
Tumor stage				0.012		0.005
pT1	15	6.2	40		40	
pT2	36	14.8	50		49.7	
pT3	177	72.8	31.7		35.6	
pT4	15	6.2	6.7		6.7	
Nodal stage				< .0001		< .0001
pN0	106	43.6	44.8		48.6	
PN1	88	36.2	33.2		34	
pN2	35	14.4	12.1		17.9	
pN3	14	5.8	0		0	
AJCC stage				< .0001		< .0001
II	125	51.4	47.6		50.8	
III	118	48.6	18.1		20.6	
Blood vessel invasion				0.033		0.003
Absent	127	52.2	39.0		41.2	
Present	116	47.8	27.2		30.7	
Differentiation				0.011		0.002
Well	4	3.4	25		25	
Moderately	106	44.1	42.5		48.2	
Poorly	133	52.5	26.5		27.1	
Postoperative RT				< .0001		< .0001
With	132	54.4	42.1		43.4	
Without	111	45.6	23.2		27.7	
Perineural invasion				0.005		< .0001
Negative	189	77.8	36.7		40.3	
Positive	54	22.2	21.7		21.7	

for patients without PORT (P = 0.138, **Figure 3A**). The 5-year OS rate were 24.8% and 18.5% for patients with and without PORT, respectively (P = 0.126, **Figure 3B**).

Among PNI-negative locally advanced ESCC patients, better outcomes were obtained in the patients received PORT. 5-year DFS and OS rates were significantly higher in patients with PORT than without PORT (46.3% and 48% vs. 24.8% and 30.7%, respectively; P < 0.001; **Figure 3C**, **3D**).

Discussion

PNI is a pathologic process characterized as tumor invading nervous structures and spreading along nerve sheaths. The pathogenesis of PNI involves complicated interactions between tumors, stromal cells and peripheral nerves. The mechanisms underlying the interactions are still far from being fully understood [13-15]. The rate of positivity for PNI is approximately 20% in colorectal cancer [4], but much higher in pancreatic cancer (80-100%) [16]. In this cohort study, 22.2% (54/243) of patients were identified as PNI positivity, of which only 18.5% (10/54) were observed at the time of resection. Similar reports by Liebig et al. [4] and Kurtz et al. [17] demonstrated that PNI was often originally underreported. The increased incidence observed on our re-evaluated resection specimen is generally because our pathologists did not routinely report PNI before.

Moreover, we showed that PNI status in ESCC was closely associated with other tumor clinic pathological characters, such as stage, infiltration depth and relapse of tumor. The positivity rate of PNI increased when the depth of invasion and pathological stage of tumor increased. In addition, we found no relationship of PNI with lymph node metastasis. Similar to our findings, previous results showed that PNI was closely correlated with the depth of invasion but not with lymph node metastases [9].

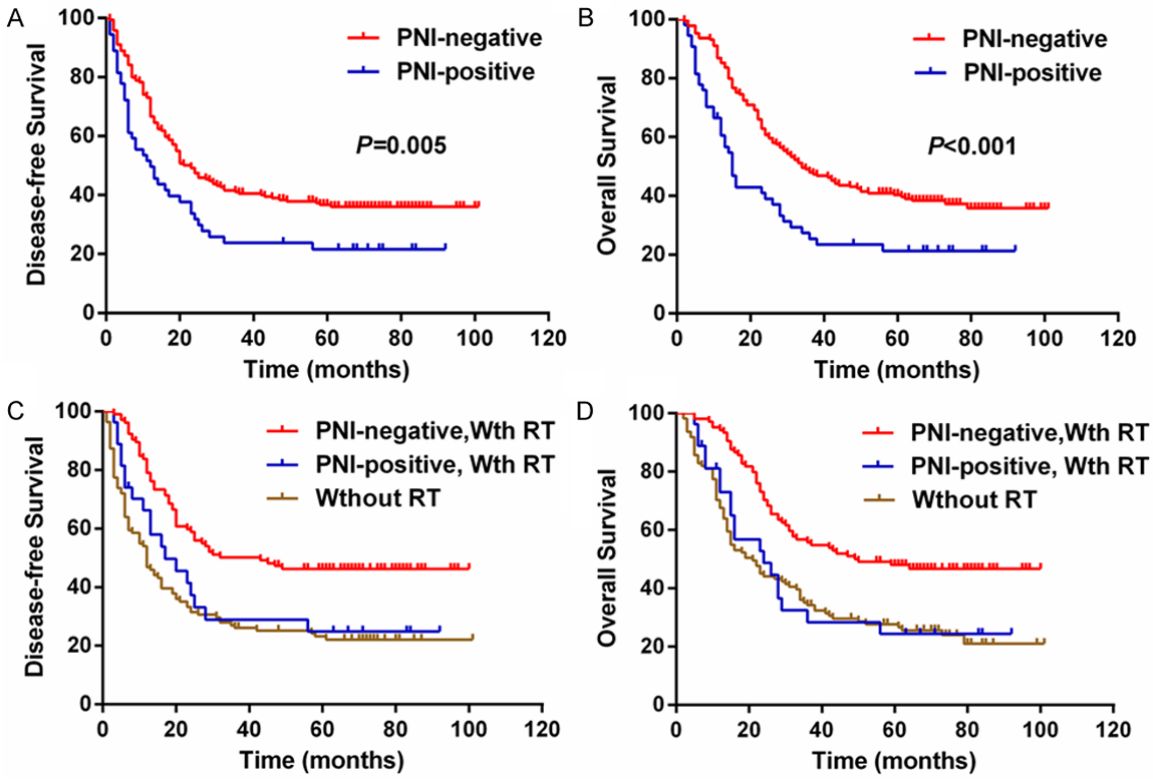

Figure 2. Kaplan-Meier curves for (A) disease-free survival (DFS) and (B) overall survival (OS) in locally advanced esophageal squamous cell carcinoma (ESCC) patients according to perineural invasion (PNI) status. Kaplan-Meier curves for (C) DFS and (D) OS among locally advanced ESCC patients based on postoperative radiotherapy (PORT) and PNI status.

However, another study reported that the positivity rate of PNI was significantly higher in patients with lymph node metastases than in patients without lymph node metastases, but no significant correlation was observed between PNI positivity and the depth of tumor invasion [18]. Recently, Tachezy et al. described that PNI was significantly correlated with advanced pT category and lymph node metastases [11]. Overall, our results further support the conclusion that PNI was significantly associated with the progression of tumor.

Local recurrence and distant metastasis are the main reasons of treatment failure for ESCC after esophagectomy. Ochiai et al. [9] showed that PNI was closely correlated with local recurrence. Local recurrence was observed in 30.0% of PNI-positive patients compared with only 4.5% of PNI-negative cases. Sarbia et al. [10] reported that incidence of PNI was significantly higher in patients with distant metastases than in patients without distant metastases. In the present study, we revealed that ESCC patients with PNI-positive are more likely to develop local recurrence and distant metastasis than PNI-negative patients. In addition, we also found that there was no relationship of the PNI positivity with lymph node metastases and blood vessel invasion. Thus, we report here that PNI is an independent spread route for the invasion and the metastasis of ESCC, and is involved not only in tumor progression, but also in tumor recurrence.

It has been demonstrated that PNI is an important predictor of aggressive tumor phenotype and poor clinical outcomes in many cancers including pancreatic cancer, head and neck cancer, prostate cancer and colorectal cancer [4, 19-21]. However, no consensus has been reached about the prognostic value of PNI in ESCC patients as yet. For instance, it was reported by several studies that ESCC patients with PNI-positive showed no significant differences in overall survival compared with PNI-negative patients [9-11]. Another study indicated that there was significant survival difference

Table 3. Cox multivariate regression analyses for the influence of PNI on overall and disease-free survival in locally advanced ESCC

Variables	Hazard Ratio	*P*	95% CI
Overall survival			
PNI	1.832	0.001	1.267-2.651
Age	1.232	0.215	0.866-1.714
Sex	0.838	0.336	0.585-1.201
Location (baseline, Proximal esophagus)			
Mid esophagus	0.924	0.808	0.489-1.745
Distal esophagus	1.028	0.937	0.519-2.037
Differentiation (baseline, Well)			
Moderately	0.840	0.447	0.193-2.064
Poorly	0.943	0.922	0.291-3.051
AJCC stage	1.949	< 0.001	1.365-2.784
Blood vessel invasion	1.124	0.512	0.793-1.592
Postoperative radiotherapy	1.743	0.001	1.263-2.404
Disease-free survival			
PNI	1.513	0.026	1.051-2.177
Age	1.088	0.612	0.785-1.508
Sex	0.832	0.384	0.794-1.818
Location (baseline, Proximal esophagus)			
Mid esophagus	0.864	0.652	0.459-1.629
Distal esophagus	0.914	0.796	0.463-1.805
Differentiation (baseline, Well)			
Moderately	0.71	0.57	0.218-2.317
Poorly	0.976	0.967	0.3-3.168
AJCC stage	1.958	< 0.001	1.376-2.787
Blood vessel invasion	1.071	0.694	0.761-1.508
Postoperative radiotherapy	1.588	0.005	1.148-2.197

between PNI-positive ESCC patients and PNI-negative ones by univariate analysis, but the independent prognostic significance of PNI was not achieved by multivariate analysis [22]. In contrast, recent study with large sample size showed that PNI was an important prognostic factor closely related to local recurrence and decreased survival in ESCC patients [18]. Moreover, Chen et al. also found PNI was an independent prognostic factor for predicting recurrence-free and overall survival in both univariate and multivariate analysis [12]. Similar to the results above mentioned, we also found that PNI, as well as other prognostic factors including tumor stage and adjuvant radiotherapy, was associated with decreased survival on multivariate analysis and indicative of an independent prognostic predictor in locally advanced ESCC patients who underwent curative resection. It needs to be mentioned that, in the previous studies, the definition of PNI don't achieve the consensus, leading to different conclusions with regard to the prognostic significance of PNI in ESCC patients.

Adjuvant radiotherapy help to kill the postoperative residual tumor foci and reduce local recurrence, but the effect of postoperative radiotherapy on survival improvement in locally advanced ESCC remains undefined [23]. It has been reported by Xiao et al. that PORT could improve the survival of N^+ or stage III esophageal cancer patients; and the 5-year survival rate was 35.1% for surgery combination with PORT in contrast to 13.1% for surgery alone [24]. However, in other randomized trials such beneficial effects on local control and OS by adjuvant radiotherapy had not been observed [25-27]. In 2004, the meta-analysis showed that no benefit has been achieved to support the use of adjuvant radiotherapy for radically resected esophageal cancer patients [28]. Taken together, it is not appropriate to recommend adjuvant radiotherapy uncritically for all locally advanced ESCC patients, as not all locally advanced ESCC patients would benefit from adjuvant radiotherapy. Here we demonstrated that the status of PNI would be helpful to the selection of patients for adjuvant radiotherapy. Our results indicated that among locally advanced ESCC patients received PORT, the prognosis of PNI-negative patients was better than that of PNI-positive patients, and there was no difference in DFS and OS between the PNI-positive locally advanced ESCC patients received PORT and locally advanced ESCC patients without PORT. Moreover, PORT didn't improve the survival in PNI-positive locally advanced ESCC patients. Locally advanced ESCC patients with PNI-negative but not PNI-positive could benefit from PORT. Overall, our results suggested that the status of PNI was indicative of high-risk ESCC phenotype with poor prognosis, and was helpful to the selection of therapeutic strategies.

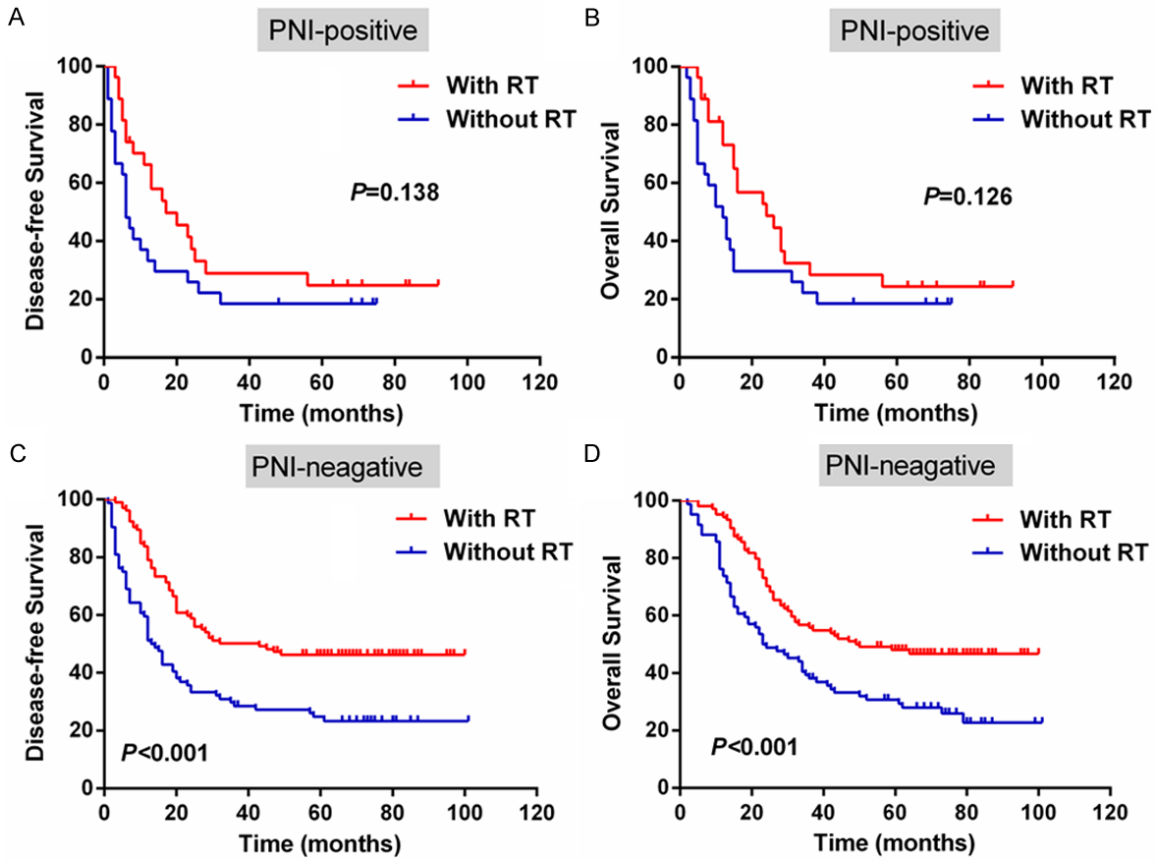

Figure 3. Kaplan-Meier curves for (A, C) disease-free survival (DFS) and (B, D) overall survival (OS) in locally advanced esophageal squamous cell carcinoma (ESCC) patients according to postoperative radiotherapy (PORT) and perineural invasion (PNI) status.

PNI is an independent spread route as blood and lymph vessel invasion, and PNI-positive patients are more likely to develop distant metastases, and interaction of tumor with peripheral nerve can make cancer cell acquire a survival and growth advantage [5, 29] and spread to distant locations beyond the scope of radiation field in the operative radiotherapy for ESCC [16]. Such factors might contribute to the poor clinical outcomes of PNI-positive patients received PORT. Previous study showed that chemoradiotherapy could reduce the incidence of PNI-positive and local recurrence in ESCC [30]. More intensive therapy, such as adjuvant concurrent chemoradiotherapy, should be suggested for this kind of high-risk patients. Furthermore, target agents against PNI in addition to chemoradiotherapy for patients with PNI-positive would be a promising treatment strategy in the future.

We recognize that there are some limitations in this study which have to be considered in the interpretation of these results. Firstly, it is a retrospective cohort study from a single-institution. It can be believed that a multicenter collaborative study with a large cohort would achieve a more convincing result. Secondly, it should be noted that only squamous cell carcinoma has been examined in this study and the results from this study may not be suitable for esophageal adenocarcinoma. Despite the limited scope of this retrospective cohort study and relatively small sample size, we believe that locally advanced ESCC patients with PNI-positive who received postoperative concurrent chemoradiotherapy may reduce the risk of recurrence and improve the clinical outcomes. Further prospective studies are needed to validate this hypothesis.

In conclusion, we demonstrated that PNI could serve as prognostic biomarker, which was significantly correlated with recurrence and poor prognosis in locally advanced ESCC after curative resection. PORT does not improve the DFS

and OS of PNI-positive locally advanced ESCC. We suggest that PORT alone should not be considered applicable to locally advanced ESCC patients with PNI-positive; for these patients intensive treatments such as postoperative concurrent chemoradiotherapy are needed.

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Disclosure of conflict of interest

None.

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References

[1] Lu F, Xue Q, Shao K, Mao YS, Gao SG, Mu JW, Tan FW, Cheng GY and He J. Preliminary experience of clinical applications of the 7th UICC-AJCC TNM staging system of esophageal carcinoma. Zhonghua Zhong Liu Za Zhi 2012; 34: 461-464.

[2] Gwynne S, Wijnhoven BP, Hulshof M and Bateman A. Role of Chemoradiotherapy in Oesophageal Cancer - Adjuvant and Neoadjuvant Therapy. Clin Oncol (R Coll Radiol) 2014; 26: 522-532.

[3] Pietras K and Ostman A. Hallmarks of cancer: interactions with the tumor stroma. Exp Cell Res 2010; 316: 1324-1331.

[4] Liebig C, Ayala G, Wilks J, Verstovsek G, Liu H, Agarwal N, Berger DH and Albo D. Perineural invasion is an independent predictor of outcome in colorectal cancer. J Clin Oncol 2009; 27: 5131-5137.

[5] Marchesi F, Piemonti L, Mantovani A and Allavena P. Molecular mechanisms of perineural invasion, a forgotten pathway of dissemination and metastasis. Cytokine Growth Factor Rev 2010; 21: 77-82.

[6] Chatterjee D, Katz MH, Rashid A, Wang H, Iuga AC, Varadhachary GR, Wolff RA, Lee JE, Pisters PW, Crane CH, Gomez HF, Abbruzzese JL, Fleming JB and Wang H. Perineural and intraneural invasion in posttherapy pancreaticoduodenectomy specimens predicts poor prognosis in patients with pancreatic ductal adenocarcinoma. Am J Surg Pathol 2012; 36: 409-417.

[7] Ueno H, Shirouzu K, Eishi Y, Yamada K, Kusumi T, Kushima R, Ikegami M, Murata A, Okuno K, Sato T, Ajioka Y, Ochiai A, Shimazaki H, Nakamura T, Kawachi H, Kojima M, Akagi Y, Sugihara K. Study Group for Perineural Invasion projected by the Japanese Society for Cancer of the C and Rectum. Characterization of perineural invasion as a component of colorectal cancer staging. Am J Surg Pathol 2013; 37: 1542-1549.

[8] Deng J, You Q, Gao Y, Yu Q, Zhao P, Zheng Y, Fang W, Xu N and Teng L. Prognostic value of perineural invasion in gastric cancer: a systematic review and meta-analysis. PLoS One 2014; 9: e88907.

[9] Ochiai M, Arai K, Funabiki T, Imazu H, Marugami Y, Futawatari H and Matsubara T. [Local spread of carcinoma of the esophagus by perineural invasion]. Nihon Geka Gakkai Zasshi 1995; 96: 137-144.

[10] Sarbia M, Porschen R, Borchard F, Horstmann O, Willers R and Gabbert HE. Incidence and prognostic significance of vascular and neural invasion in squamous cell carcinomas of the esophagus. Int J Cancer 1995; 61: 333-336.

[11] Tachezy M, Tiebel AK, Gebauer F, Kutup A, Tharun L, Pantel K, Izbicki JR and Vashist YK. Prognostic impact of perineural, blood and lymph vessel invasion for esophageal cancer. Histol Histopathol 2014; 29: 1467-75.

[12] Chen JW, Xie JD, Ling YH, Li P, Yan SM, Xi SY, Luo RZ, Yun JP, Xie D and Cai MY. The prognostic effect of perineural invasion in esophageal squamous cell carcinoma. BMC Cancer 2014; 14: 313.

[13] Liebig C, Ayala G, Wilks JA, Berger DH and Albo D. Perineural invasion in cancer: a review of the literature. Cancer 2009; 115: 3379-3391.

[14] Chedotal A, Kerjan G and Moreau-Fauvarque C. The brain within the tumor: new roles for axon guidance molecules in cancers. Cell Death Differ 2005; 12: 1044-1056.

[15] Cornell RJ, Rowley D, Wheeler T, Ali N and Ayala G. Neuroepithelial interactions in prostate cancer are enhanced in the presence ofprostatic stroma. Urology 2003; 61: 870-875.

[16] Bapat AA, Hostetter G, Von Hoff DD and Han H. Perineural invasion and associated pain in pancreatic cancer. Nat Rev Cancer 2011; 11: 695-707.
[17] Kurtz KA, Hoffman HT, Zimmerman MB and Robinson RA. Perineural and vascular invasion in oral cavity squamous carcinoma: increased incidence on re-review of slides and by using immunohistochemical enhancement. Arch Pathol Lab Med 2005; 129: 354-359.
[18] Tanaka A, Matsumura E, Yosikawa H, Uchida T, Machidera N, Kubo R, Okuno K, Koh K, Watatani M and Yasutomi M. An evaluation of neural invasion in esophageal cancer. Surg Today 1998; 28: 873-878.
[19] Chen TC, Wang CP, Ko JY, Yang TL, Hsu CW, Yeh KA, Chang YL and Lou PJ. The impact of perineural invasion and/or lymphovascular invasion on the survival of early-stage oral squamous cell carcinoma patients. Ann Surg Oncol 2013; 20: 2388-2395.
[20] Hirai I, Kimura W, Ozawa K, Kudo S, Suto K, Kuzu H and Fuse A. Perineural invasion in pancreatic cancer. Pancreas 2002; 24: 15-25.
[21] D'Amico AV, Wu Y, Chen MH, Nash M, Renshaw AA and Richie JP. Perineural invasion as a predictor of biochemical outcome following radical prostatectomy for select men with clinically localized prostate cancer. J Urol 2001; 165: 126-129.
[22] Torres CM, Wang HH, Turner JR, Richards W, Sugarbaker D, Shahsafaei A and Odze RD. Pathologic prognostic factors in esophageal squamous cell carcinoma: a follow-up study of 74 patients with or without preoperative chemoradiation therapy. Mod Pathol 1999; 12: 961-968.
[23] Berger B and Belka C. Evidence-based radiation oncology: oesophagus. Radiother Oncol 2009; 92: 276-290.
[24] Xiao ZF, Yang ZY, Liang J, Miao YJ, Wang M, Yin WB, Gu XZ, Zhang DC, Zhang RG and Wang LJ. Value of radiotherapy after radical surgery for esophageal carcinoma: a report of 495 patients. Ann Thorac Surg 2003; 75: 331-336.
[25] Teniere P, Hay JM, Fingerhut A and Fagniez PL. Postoperative radiation therapy does not increase survival after curative resection for squamous cell carcinoma of the middle and lower esophagus as shown by a multicenter controlled trial. French University Association for Surgical Research. Surg Gynecol Obstet 1991; 173: 123-130.
[26] Fok M, Sham JS, Choy D, Cheng SW and Wong J. Postoperative radiotherapy for carcinoma of the esophagus: a prospective, randomized controlled study. Surgery 1993; 113: 138-147.
[27] Zieren HU, Muller JM, Jacobi CA, Pichlmaier H, Muller RP and Staar S. Adjuvant postoperative radiation therapy after curative resection of squamous cell carcinoma of the thoracic esophagus: a prospective randomized study. World J Surg 1995; 19: 444-449.
[28] Malthaner RA, Wong RK, Rumble RB, Zuraw L and Gastrointestinal Cancer Disease Site Group of Cancer Care Ontario's Program in Evidence-based C. Neoadjuvant or adjuvant therapy for resectable esophageal cancer: a clinical practice guideline. BMC Cancer 2004; 4: 67.
[29] Ayala GE, Dai H, Ittmann M, Li R, Powell M, Frolov A, Wheeler TM, Thompson TC and Rowley D. Growth and survival mechanisms associated with perineural invasion in prostate cancer. Cancer Res 2004; 64: 6082-6090.
[30] Tanaka A, Ishikawa S, Okuno K and Shiozaki H. Effect of chemoradiotherapy for neural invasion in an advanced esophageal cancer. Hepatogastroenterology 2008; 55: 963-966.