

Evaluation of risk factors for lymph node metastasis in T2 lower rectal cancer to perform chemoradiotherapy after local resection

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Abstract. The oncological outcome of chemoradiotherapy (CRT) after local excision (LE) for T2 lower rectal cancer has demonstrated a high local recurrence (LR) rate. The aim of the present study was to determine the risk factors for lymph node metastasis (LNM) in order to reduce LR in T2 lower rectal cancer after LE and CRT. Specimens were collected from 95 consecutive patients with T2 lower rectal adenocarcinoma who underwent R0 resection by total mesenteric excision or tumor-specific mesenteric excision between January 2008 and December 2018 at Osaka International Cancer Institute. All specimens were checked and evaluated to determine the risk factors for LNM. LNM was observed in 26 patients (27%), including 2 patients (2%) with lateral pelvic lymph node metastasis. Univariate analysis indicated lymphovascular invasion (LVI; $P=0.008$), tumor budding ($P=0.012$) and histology other than well-differentiated adenocarcinoma ($P=0.08$) were associated with LNM; multivariate analysis revealed that LVI ($P=0.03$) was the only independent risk factor for LNM. LNM was confirmed in 0% (0/8) of patients without LVI, tumor budding and histological type. LVI, tumor budding and histological type can be risk factors for LNM in lower rectal cancer. The present study may be helpful to select patients for performing LE and CRT with good oncological outcome.

Introduction

Generally, additional total mesenteric excision (TME) is recommended for the treatment of pathological T2 lower rectal cancer after local excision (LE) because the high risk for local recurrence (LR) persists even if LE was performed

curatively (1). Although TME is the gold standard procedure for lower rectal cancer, this procedure is associated with complications such as sexual, urinary, or anal dysfunction, and with temporary or permanent stoma. In particular, anal dysfunction and stoma can affect patients' quality of life (2,3) and are serious problems for the elderly and those who desire anal preservation. Recently, combination treatment of postoperative chemoradiotherapy (CRT) after LE for T1/T2 rectal cancer has become one optional treatment in the National Comprehensive Cancer Network guidelines (4), and several studies have shown good oncological outcomes and excellent anal function compared to TME, particularly in T1 rectal cancer (5-8). However, a few studies on long-term patient outcomes in T2 rectal cancer showed LE to have a higher LR rate than TME (5,7,8). Hence, when undertaking postoperative CRT after LE in T2 rectal cancer, it is necessary to select patients with a low risk of recurrence, and in particular it is useful to select patients with a low risk of lymph node metastasis (LNM) (9-11). Although some reports showed risk factors of nodal involvement in T2 colorectal cancer (9-13), a few reports limited to lower rectal cancer had indications for LE. The aim of this study was to evaluate the risk factors of nodal involvement with updated parameters in T2 lower rectal cancer and to select patients with low risk of LNM.

Materials and methods

Study design and patient characteristics. This is a retrospective cohort study at Osaka International Cancer Institute. The study protocol was approved by the institutional review board (No. 18033-2). Written informed consent was waived because of the retrospective design. We collected specimens from 95 consecutive patients with pathological T2 lower rectal adenocarcinoma who underwent R0 resection by TME or TSME (14) between January 2008 and December 2018 (Table I). Exclusion criteria were recurrent carcinoma, multiple carcinoma, stage IV, and preoperative chemotherapy or radiotherapy. To evaluate rectal cancer which has indication for LE, we picked up rectal cancer located within 10 cm from the anal verge as lower rectal cancer.

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Surgical procedure. TME/TSME was performed with D3 dissection (15). Bilateral lateral pelvic lymph node dissection was performed after TME/TSME for clinical T3 tumors.

Pathology evaluation. After formalin fixation, more than two pathologists cut the rectum and lymph nodes along the long axis, and performed microscopic examinations of hematoxylin and eosin stained specimens. A pathologist (M.K) who was blind to the clinical information reviewed all tumors on silane-coated glass slides. Clinicopathological data are shown in Table II. A desmoplastic reaction (DR) was defined as a fibrotic stromal response in cancer development and was divided into three groups: Mature, when the stroma was composed of mature collagen fibers; intermediate, when keloid-like collagen was intermingled with mature fibers; and immature, consisting of a myxoid stroma that included no mature fibers (16). To evaluate the association between LNM and the extent of tumor invasion in the muscularis propria (MP) layer, the depth/width of tumor invasion into the MP layer were measured, and the tumor rate of invasion into MP layer was calculated by dividing the length of tumor invasion into the MP layer by the thickness of the MP layer.

Statistical analysis. The parameters of tumor size, depth of tumor invasion into the MP layer, width of tumor invasion into the MP layer, and rate of invasion into the MP were evaluated by receiver operating characteristic (ROC) curves using the area under the curve (AUC) to determine the optimal cut-off value. The associations between LNM and these parameters were analyzed with the χ^2 test. Variables with a P-value <0.2 in a univariate analysis were further evaluated in a multivariate logistic model. Three-year-relapse free survival was determined using the Kaplan-Meier method. Values of P<0.05 were considered statistically significant. Statistical analyses were performed using JMP version 8.0.2 software (SAS Institute, Cary, NC).

Results

Clinicopathological characteristics. The demographic and pathological characteristics of patients are provided in Table I. The median follow-up duration was 37.4 months (range 1.0-120.1). LNM was confirmed in 26 patients (27%), including 2 patients (2%) with lateral pelvic lymph node metastasis. Fourteen patients developed recurrence, of whom 5 had local recurrence, 6 had distant metastases and 3 had both local and distant metastases.

Parameters and associations between parameters and LNM. The AUC for LNM and cut-off values for tumor size and depth, width, and rate of invasion into the MP layer were 0.61 and 23 mm, 0.51 and 2 mm, 0.55 and 12 mm, and 0.49 and 80%, respectively. Univariate analyses of the association between these parameters and LNM are represented in Table II. LVI (P=0.008), and tumor budding (P=0.012) were significantly related to LNM, and a possible association between LNM and histology other than well-differentiated adenocarcinoma was indicated (P=0.083); however, there were no relationships between LNM and these updated parameters, such as depth, width, and rate of invasion into the MP layer, based on the

Table I. Clinicopathological characteristics.

Characteristics	All patients (n=95)
Age, years (range)	70 (33-84)
Sex (male:female), n	59:36
Distance of tumor from AV, cm	
10>X>5	51
5≥X	44
pN0/pN1, n	69/26
Lateral pelvic lymph node metastasis, n	2
pStage I/IIIA/IIIB, n	69/21/5
Recurrence, n	14
3-year RFS, %	84.9

AV, anal verge; RFS, relapse-free survival.

ROC curves and the AUC values and also state (data not shown).

Risk factors for LNM. Multivariate analysis revealed that LVI (P=0.03) was an independent risk factor for LNM. Histology also showed a tendency toward being a risk factor, but there was no significant difference (P=0.07) (Table III). Table IV shows the rate of LNM in relation to these risk factors of LVI, tumor budding, and histological type. LNM was confirmed in 9 patients (9/13, 69.2%) with three risk factors, in 10 patients (10/39, 29.6%) with two risk factors, and in 7 patients (7.34, 20.6%) with one risk factor. LNM was not confirmed in any patients lacking all 3 risk factors (0/9).

Discussion

This study investigated the relationship between LNM and clinicopathological parameters in T2 lower rectal cancer for select patients with a low risk of LNM, and revealed LVI, tumor budding, and histology other than well-differentiated adenocarcinoma were risk factors for LNM. The values of our study are limited to lower rectal cancer, which had indications for LE, and evaluating updated parameters such as DR, tumor budding, perineural invasion, and the extent of tumor invasion into the MP (9,12,17,18).

This paper showed that LVI was an independent risk factor for LNM in T2 lower rectal cancer by multivariate analysis. In addition, LVI and tumor budding were risk factors and histological type was likely to be a risk factor for LNM in T2 lower rectal cancer by univariate analysis. These results were similar to a previous study by Kobayashi *et al* (11) published in 2010, which showed biological sex, histology other than well-differentiated adenocarcinoma, and lymphatic invasion as risk factors for LNM in lower rectal cancer in a large retrospective multi-institutional study. Kobayashi *et al* (11) showed that the LNM rate in T2 patients was 9.1% without lymphatic invasion; however, our data revealed no patients (0/9) without unfavorable pathological features who had risk of LNM (Table IV), even though the number was small by

Table II. Univariate analysis for factors associated with lymph node metastasis.

Factors	LN negative, (n=69)		LN positive (n=26)		P-value
	Number	%	Number	%	
Age, years					0.246
<65	28	66.6	14	33.4	
≥65	41	77.3	12	22.7	
Sex					0.622
Male	44	74.6	15	25.4	
Female	25	69.4	11	30.6	
AV, cm					0.934
≥5	46	71.9	18	28.1	
<5	23	74.2	8	25.9	
Morphology					0.692
Polypoid	13	76.5	4	23.5	
Non-polypoid	56	71.8	22	28.2	
Size, mm					0.117
≥23	62	75.6	20	24.4	
<23	7	53.8	6	46.2	
Histology					0.083
Well-differentiated	23	33.3	4	15.4	
Others	46	66.7	22	84.6	
Lymphovascular invasion					0.008
Negative	25	89.3	3	10.7	
Positive	44	65.7	23	34.3	
Tumor budding					0.012
Grade 1	61	78.2	17	21.8	
Grade 2/3	8	47.1	9	52.9	
Perineural invasion					0.175
Negative	63	75.0	21	25.0	
Positive	6	54.5	5	45.5	
Desmoplastic reaction					0.211
Mature	28	80.0	7	20.0	
Intermediate/immature	41	68.3	19	31.7	
Depth of invasion into MP layer, mm					0.854
<2	28	73.7	10	26.3	
≥2	41	71.9	16	28.1	
Width of invasion into MP layer, mm					0.122
<12	38	66.7	19	33.3	
≥12	30	81.1	7	18.9	
Rate of invasion into MP layer, %					0.247
<80	64	74.4	22	25.6	
≥80	5	55.6	4	44.4	

AV, anal verge; LN, lymph node; MP, muscularis propria.

univariate analysis. Hence, we considered these risk factors may be more valuable indicators to select an LNM-negative patient group. Similar to previous reports (9-11,16-18), the present study also included clinical LNM-positive patients. If they are excluded on CT or MRI in clinical practice, patients with favorable pathological features are expected to

have a lower risk of LNM. In general, a poorly differentiated component was recognized as one of the risk factors for LNM (9,10,17,19,20). However, the present study included only 2% of poorly differentiated adenocarcinoma; therefore, we did not statistically analyze this risk factor. Table V showed some risk factors published in previous reports,

Table III. Multivariate analysis for factors associated with lymph node metastasis.

Variables	OR	95% CI	P-value
Size (≥ 23)	0.42	0.10-1.81	0.24
Histology	2.95	0.89-11.90	0.07
Lymphovascular invasion	4.00	1.13-19.10	0.03
Tumor budding	1.99	0.49-7.93	0.32
Perineural invasion	1.16	0.24-5.18	0.84
Length of invasion in MP	0.47	0.15-1.36	0.17

OR, odds ratio; MP, muscularis propia.

including updated data such as the extent of tumor invasion into the MP (12,17,18), DR (9), and perineural invasion (10). However, in our study these factors did not show statistically significant differences.

Garcia-Aguilar *et al* (1) showed a high LR rate (37%, n=27) by LE alone, even if the study involved patients with good pathological features (i.e., without LVI and mucinous component and with R0 resection). They indicated additional CRT should be adopted to control LR. Some recent reports have shown a high LR rate even after LE and CRT (5,7,8). Greenberg *et al* (8) showed 18% LR rate (n=51) that included patients with unfavorable pathological features who were clinical LNM positive. Rackley *et al* (5) showed a 22% LR rate (n=42) that included patients with unfavorable pathological features, positive margin, and unevaluable margin after LE, although patients that were clinically LNM positive were excluded. Considering these findings, to perform LE and CRT for T2 lower rectal cancer with a good oncologic outcome, it is necessary to select patients with

Table IV. Rate of LNM depending on risk factors.

Number of risk factors	Patients, n	LNM patients, n	Rate of LNM, %
3	13	9	69.2
2	39	10	25.6
1	34	7	20.6
0	9	0	0

Risk factors: Lymphovascular, tumor budding, histological type. LNM, lymph node metastasis.

at least good pathological features and R0 local resection by LE, and without clinical LNM.

Some limitations must be considered when interpreting the results of this study. First, this study was a single-institute, retrospective analysis. Second, because this study was small, we chose the histological type as a marginally significant risk factor that showed $P < 0.1$ in univariate analysis, in addition to the two other factors, and we analyzed the rate of LNM with the risk factors calculated by univariate analysis (Table IV). Third, this study showed the risk factors for LNM with specimens after TME. When adopting these risk factors for LE, we should recognize that pathological evaluation may become insufficient, for example due to heat degeneration around the tumor. Fourth, in the clinical setting, we have to perform local excision instead of biopsy to obtain information about the risk factors such as LV1 and tumor budding.

In conclusion, our study showed LVI, tumor budding, and histological type can be risk factors for LNM in lower rectal cancer. When performing LE and CRT for T2 rectal cancer

Table V. Literature reports about the risk factors for lymph node metastasis in colorectal cancer.

Author, year	Location	T1	T2	Risk factors in T2	(Refs.)
Kajiwara <i>et al</i> , 2010	Colorectal	-	244	Female, LVI, tumor budding, poorly differentiated component, myxoid cancer stroma, extent of tumor invasion in MP	(9)
Masaki <i>et al</i> , 2005	Rectum	-	72	Female, tumor budding	(13)
Kobayashi <i>et al</i> , 2010	Lower rectum	233	334	Female, lymphatic invasion, histological type	(11)
Tong <i>et al</i> , 2011	Colorectal	-	317	LVI, extent of tumor invasion in MP	(17)
Ding <i>et al</i> , 2011	Rectum	-	346	Age, location, extent of tumor invasion in MP, histological type	(18)
Chang <i>et al</i> , 2012	Rectum	264	679	LVI, histological type, morphology, perineural invasion, desmoplasia (T1/T2),	(10)
Saraste <i>et al</i> , 2013	Rectum	205	472	LVI, histological type (T1/T2)	(19)
He <i>et al</i> , 2015	Rectum	113	463	Histological type, extent of tumor invasion in MP, CA19-9, ulcerative mass, (T1/T2)	(12)
Yoshida <i>et al</i> , 2018	Colorectal	202	115	Location, tumor size, lymphatic invasion, histological type	(20)
Present study, 2019	Lower rectum	-	95	LVI, tumor budding, histological type	-

LVI, lymphovascular invasion; MP, muscularis propia.

based on risk factors, a large prospective multicenter study should be conducted.

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Availability of date and materials

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

Authors' contributions

HU, MO, MY and ST conceived and designed the present study. HU and MO wrote the manuscript. MK and SN were responsible for the pathological examination. NH, JN, MY, HW, HT, TO and HM performed follow-up and collected the data. MY and ST revised the manuscript critically for important intellectual content. All authors read and approved the final manuscript.

Ethics approval and consent to participate

This was a retrospective cohort study at Osaka International Cancer Institute. The study protocol was approved by the Institutional Review Board of the Osaka International Cancer Institute (approval no. 18033).

Patient consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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