

# The Prognostic Significance of Number of Lymph Node Metastasis in Colon Cancer – Based on Japanese Techniques of Resection and Handling of Resected Specimens

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## 1. Introduction

Staging systems for cancer reflects the prognosis of the disease and it is used to choose the modality of treatment. The TNM classification has mainly been used in the west. In Japan, Japanese classification according to General Rules for Clinical and Pathological Studies on Cancer of the Colon, Rectum and Anus(JGR) (Japanese Society for cancer of colon and rectum, 2009) is used. The degree of the lymph node metastasis in each staging system has some variations.

In 2009, 7th edition of (JGR, 2009) was revised to make it uniform with the 6<sup>th</sup> edition of TNM classification (Sobin & Wittekond, 2002). However, the 7<sup>th</sup> edition of TNM classification (Sobin et al., 2009) was further revised where the category of nodal status was subdivided (Table 1) on the basis of number of positive lymph nodes. The validity of which is based on the pooled SEER database of 109,953 cases of colorectal cancer lymph node metastases (Gunderson et al., 2010). Japanese classification of nodal status takes into account not only the number of positive lymph nodes but also the site from where they are retrieved according to the location of the tumor. Our study showed recategorization of lymph nodes such as 1, 2 to 6 and 7 or more lymph nodes with metastasis reflected the prognosis of the disease (Akagi et al., 2010). Thus, the number of lymph nodes retrieved plays a vital role in the staging system and is one of the main prognostic indicators of the disease. The various techniques of resection and handling of resected specimens may also vary according to different institutions and countries. The number of lymph nodes retrieved can depend on different factors like the surgical technique, length of resection, mesocolic excision, lymph node dissection, handling of resected specimen and criteria for pathological diagnosis which has some differences in Japan as compared to the west. Moreover, chemotherapy protocols and treatment of recurrence also may vary in different places. This can alter the stage, recurrence rate, as well as the outcome of the disease. Therefore, here we have elaborated our technique of resection, specimen handling and nodal dissection which is uniformly practiced in all centers of Japan and present data from our center where these techniques have been carried out consecutively.

		6th				7th	
		LN category		stage			stage
TNM	N1	1-3 regional LNM	IIIA~ IIIB	⇒	N1a	1 regional LNM	IIIA~ IIIC
					N1b	2-3 regional LNM	
					N1c	Satellite without regional nodes	
	N2	4 or more regional LNM	IIIC		N2a	4-6 regional LNM	IIIB~ IIIC
					N2b	7 or more regional LNM	
JGR	N1	Metastasis in pericolic LN	IIIA	⇒	N1	1-3 pericolic/perirectal, intermediate LNM	IIIA
	N2	Metastasis in intermediate LN	IIIB		N2	4 or more pericolic/perirectal, intermediate LNM	IIIB
	N3	Metastasis in main LN			N3	Main LNM (include Lateral LN)	
	N4	Metastasis in paraaortic LN	IV		M1	Metastasis beyond regional LN	IV

LN; Lymph node, LNM; Lymph node metastasis, JGR - Japanese classification according to General Rules for Clinical and Pathological Studies on Cancer of the Colon, Rectum and Anus

Table 1. The changes of lymph nodes category between former and current system

## 2. Methods

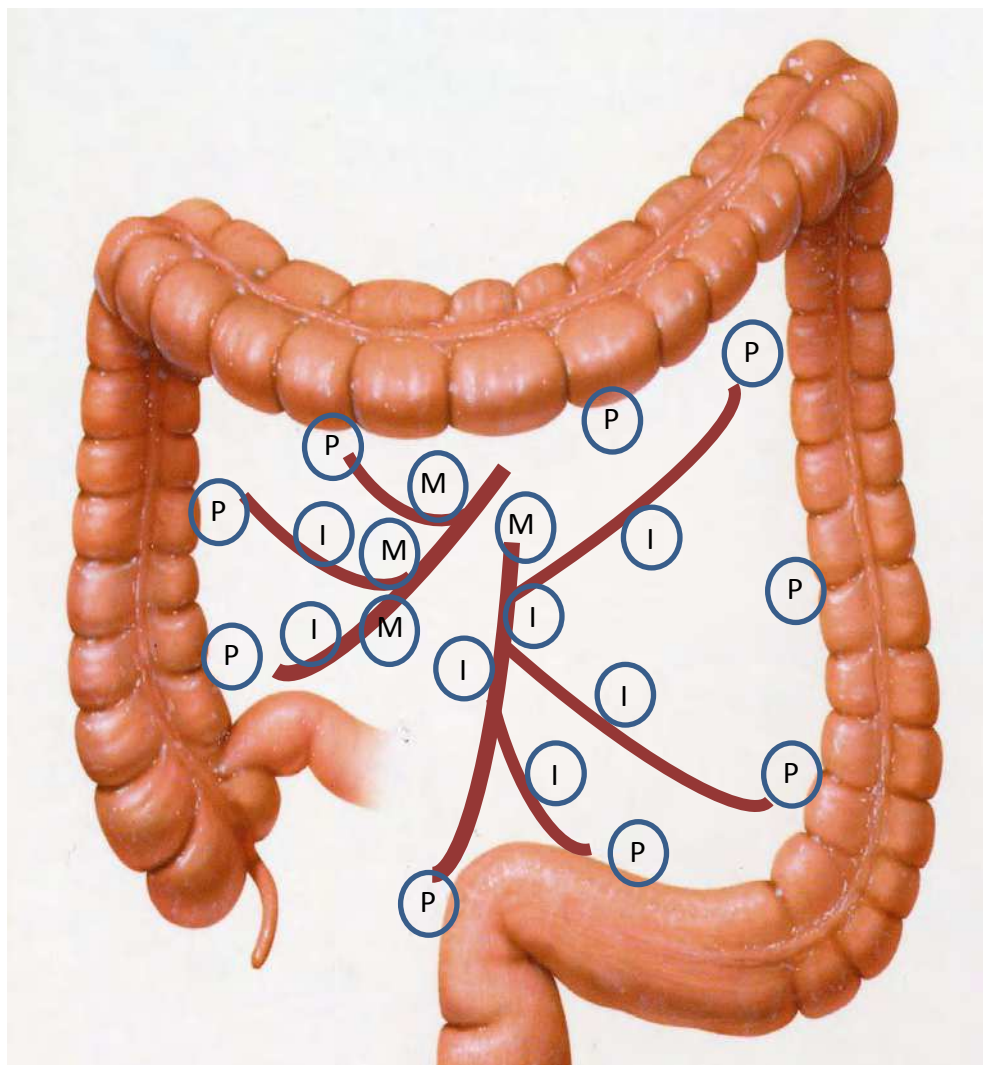
### 2.1 Patients

A total of 1107 patients with primary colon cancer treated by curative resection from January 1, 1985 to December 31, 2006, were identified from the colorectal cancer database of Kurume University, Fukuoka, Japan. Of these patients, 361 patients with Dukes C colon cancer located from caecum to recto sigmoid junction were included in this study. Patients who underwent neoadjuvant chemo-radiotherapy, patients with familial adenomatous polyposis (FAP) or inflammatory bowel disease (IBD) and patients with rectal cancer were excluded from the study. The median age of patients was 66 years (64.9±12.6) and 213 (59%) patients were male. Almost all patients were administered oral prodrug of 5-fluorouracil as postoperative adjuvant chemotherapy. The median number of nodes examined was 28 (range, 5-108, average, 30.7 ±16.5) and the median duration of follow-up was 68 months (24-186 months, average, 60.5±22.3) from the date of their initial surgery.

### 2.2 Surgical technique for resection of colon and staging of lymph node status







Surgery for colorectal cancer was performed by only certified colorectal surgeons. A similar protocol for length of resection and lymph node dissection for colon cancer was followed by all surgeons. The extent of the resection was determined by the location of cancer, its feeding arteries, cancer staging and the pattern of potential lymphatic spread. The feeding arteries were superior and inferior mesenteric artery and its branches such as ileocolic, right colic, middle colic, left colic and sigmoid arteries. The regional lymph nodes consisted of three groups; main, intermediate and pericolic lymph nodes (Figure 1.) Complete mesocolic

excision by sharp dissection of the entire mesocolon with intact facial layers and ligation of the supplying vessels at its origin was performed (Hohenberger et al. 2009). The pedicle of artery of the main lymph nodes was ligated and cut. In principle, the extent of mesocolon supplied by the feeding artery and all regional lymph nodes were removed en block for advanced cancer (Table 2.). For early cancer, the intermediate and pericolic lymph nodes of feeding artery were removed. Length of bowel resection was 10cm proximally and distally from location of the feeding arteries where the tumor was located.



M : Main lymph nodes, I : Intermediate lymph nodes P : Pericolic lymph nodes

Fig. 1.

	Surgical procedures	Ligation arteries and (lymphadenectomy)
	Ileocecal resection	Ileocolic artery (Ileocolic root nodes)
	Right hemicolectomy	Rt.colic artery (Rt.colic root nodes)
	Transverse colectomy	Middle colic artery (Mid.colic root nodes)
	Left colectomy	Lt. colic artery (Inferior mesenteric nodes)
	Sigmoidectomy	Inferior mesenteric artery (Inferior mesenteric nodes)
	Anterior resection	Inferior mesenteric artery (Inferior mesenteric trunk nodes)

The intestine with tumor and adjacent mesocolon (▲: Lymph nodes are included in this) removed after ligation of pedicle (●).

Table 2. Operation for advanced colon cancer

### 2.3 Handling of resected specimen

Lymph node dissection was carried out by the surgeon prior to formalin fixation in fresh resected specimens. The lymph nodes along the feeding vessels were picked up from the mesocolon and kept separately according to the lymph node stations and fixed in formalin (Fig. 2a,b,c). The pericolic nodes in the fat tissue beside the tumor were left intact for the correct judgment of depth of invasion. The opened intestine was placed on a board with the mucosal side up and the edge stretched and pinned to reproduce its original appearance. After formalin fixation for several days the tumor was sectioned at 5 mm intervals (Fig. 2d). One of the deepest invasive specimens was examined by expert pathologist (Fig. 2e). The final decision of histological examination of specimen and lymph node metastasis was made by the surgical colorectal pathologist (K. Shirouzu; co-author).

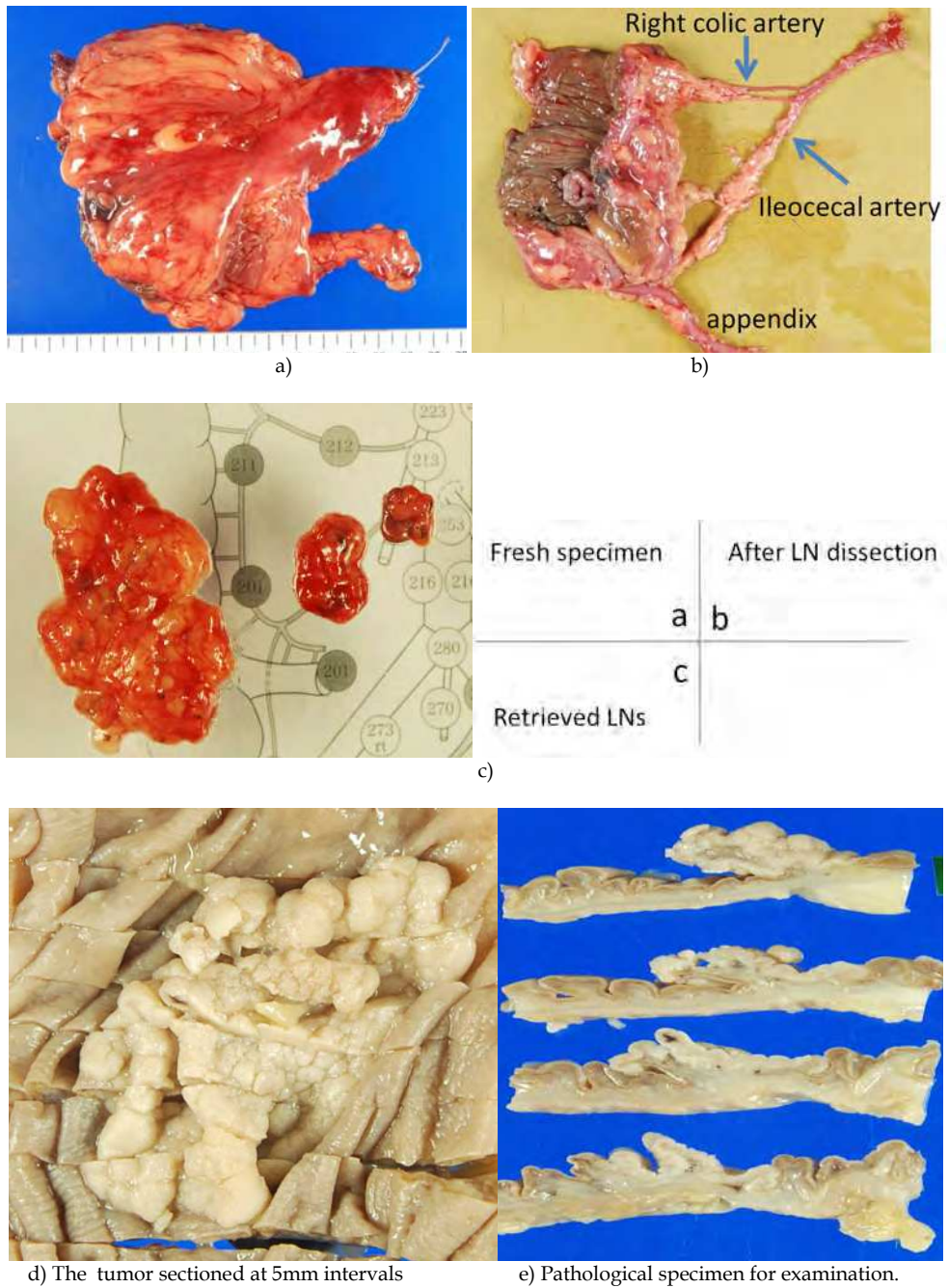


Fig. 2. Handling of fresh specimen after ileocecal resection

## 2.4 Statistical analysis

The cases were classified according to the number of metastatic lymph nodes. Survival rate for each group was assessed. A new classification was then considered to recategorize the lymph nodes from cases with similar survival rates. In the new classification, survival rate was assessed with prognostic-relate factors inferred statistically. Analysis of variance or a t-test was used to analyze continuous variables.  $\chi^2$  test was used for categorical variables. Five-year survival rates and prognostic factors were estimated using Kaplan-Meier survival method and Cox proportional hazard regression model, respectively. Log-rank test was used to assess whether survival differences were significant. Values of  $p < 0.05$  was considered statistically significant. Statistical analysis was performed using JMP software ver. 8.0 (SAS Institute, Cary, NC, USA).

## 3. Results

### 3.1 Number of lymph nodes retrieved and Lymph Node Metastasis (LNM)

Table 3. shows the number of cases, average number of lymph nodes retrieved, five year overall survival and recurrence rate in each group divided by number of LNM. The most common was the group which had one LNM. The median number of LNM was three (range, 1-14). The average number of lymph nodes retrieved in each group was from 26 to 47, and there is significant relation between number of LNM and lymph nodes retrieved. The more the number of lymph nodes retrieved increased, the number of LNM also increased. Similarly the recurrence rate was higher when the number of lymph nodes retrieved was more. However, it did not show statistical significance. The 5 year survival rate for each group classified by the number of LNM is shown in Table 3. 5-year survival rate of the group with 1 LNM was significantly better than that of other groups and the group with  $\geq 7$  LNM showed significantly worst survival than groups with  $\leq 6$  LNM.

Number of LNM*	Case	Ave. of LN s retrieved*/**	recurrence rate**	5 yr OS		5 y s.OS	
1	143	26 (5-108)	21.7	81.6%	⇒	81.6%	
2	68	26 (5-66)	23.9	72.2%		} ⇒	70.9%
3	53	28 (5-70)	28.3	71.4%			
4	33	31 (6-78)	24.2	68.9%			
5	19	29 (19-71)	31.6	68.1%			
6	11	40 (17-71)	18.2	70.0%		} ⇒	44.1%
7	8	33 (12-52)	37.5	46.9%			
8	6	38 (32-68)	50	42.6%			
9	10	45 (17-86)	50	48.9%			
10 or more	14	47 (21-70)	57.1	29.9%			

LNM: Lymph nodes metastasis, Ave.: average (range), 5 yr OS: 5 year overall survival

\*:  $p < 0.001$  (number of LNM vs Ave. of LNs retrieved,

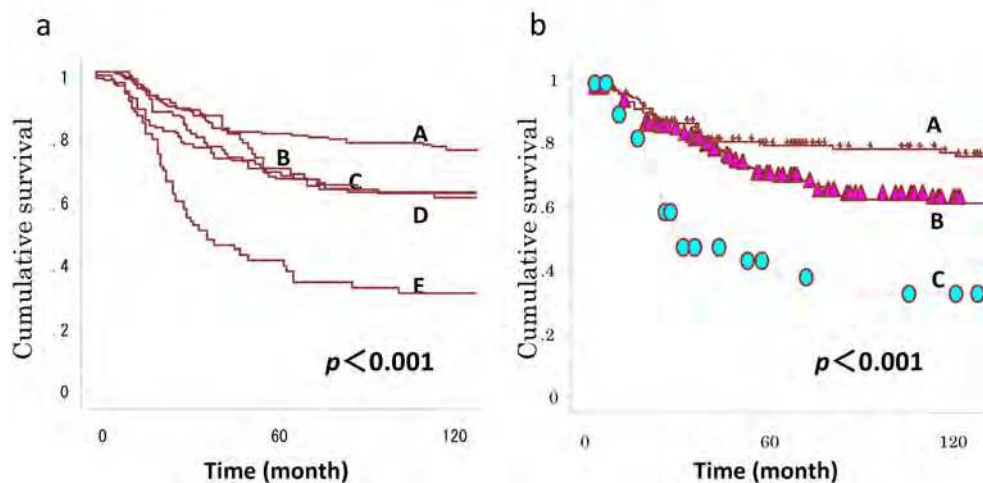
\*\* :  $p = 0.763$  (Ave. of LNs retrieved vs recurrence rate)

Table 3. Recurrence and Survival on the number of lymph nodes metastasis



### 3.2 Recategorization and its association with survival

The survival curves of five groups based on the former method (Akagi et al., 2010) were compared. Group A consisted of cases of 1 LNM, group B 2 LNM, group C 3 LNM, group D 4-6 LNM, and group E 7 or more LNM. Survival curves for group B, C and D were similar (Fig. 3a). Based on the above-mentioned results, the survival curve of each group, survival rate, patient number and current classification was considered and integrated and reorganized into three groups (Table 3, Figure 3b). The new classification used 1 LNM for group A (143 cases), 2-6 LNM for group B (184 cases), and  $\geq 7$ LNM for group C (38 cases). In brief, N category seemed the most appropriate when the number of LNM was classified as 1, 2-6, and  $\geq 7$ . The 5-year survival rate for each group was A; 81.6%, B; 70.9%, and C; 44.1%, respectively (Table 3). This classification more accurately reflected the prognosis as compared to the conventional categories for LNM. Factors influencing prognosis were extracted from clinicopathological factors of every group by univariate analysis. Then, correlative prognostic factors were included in the new classification of LNM degree was estimated with multivariate analysis (Table 4). As for independent prognostic factors, degree of venous invasion and seven and more LNM were considered to be poor prognostic factors.



(a) According to number of lymph node metastasis (LNM).  
 A: 1 LNM, B: 2 LNM, C: 3 LNM, D: 2-6 LNM, E: 7 and more LNM  
 (b) According to recategorized group  
 A: 1 LNM, B: 2-6 LNM, C: 7 or more LNM

Fig. 3. Cumulative survival curves

Variable	Univariate		Multivariate		
	<i>p</i> -value(logrank test)	HR (95%CI:L-U)	<i>p</i> -value	Chi-square	
Number of LN metastasis					
≥ 7 vs. ≤ 6	0.0001	2.11 (0.137–1.310)	0.0174	5.66	
Depth of invasion					
se vs. ss	0.0076	1.76 (-0.014–1.224)	0.0562	3.65	
Histologic type					
poor,muci vs. well,mod.	0.0952	1.13 (-0.539–0.731)	0.7019	0.15	
Lymphatic invasion					
ly2-3 vs. ly0-1	0.0025	1.46(-0.205–0.935)	0.1972	1.67	
Venous invasion					
v2-3 vs. v0-1	0.0013	2.63 (0.187–1.628)	0.0175	5.65	
Preoperative CEA value					
≥ 5 vs. <5	0.1751	1.15 (-0.349–0.554)	0.0673	0.22	

LN: lymph node, se: serosa, ss: sub serosa, poor: poorly differentiated adenocarcinoma, mucic: mucinous carcinoma, well: well differentiated adenocarcinoma, mod: moderately differentiated adenocarcinoma, ly0-1, v0-1: negative to minimal invasion, ly2-3, v2-3: moderate to severe invasion, CEA:carcinoembryonic antigen, CI, confidence interval

Table 4. Independent Prognostic Factor for Disease Specific Survival using Cox Regression Analysis

#### 4. Discussion

Depth of invasion and number of regional lymph node metastasis(LNM) are known to be important prognostic factors for colorectal cancer, and these factors are used to determine the stage of the disease (Chapuis et al.,1985; Vaccaro et al., 2004; Choen et al., 1991). In Japan, on the basis of clinical studies on colorectal cancer the general rules for clinical and pathological studies of cancer of the colon, rectum and anus which has been modified continuously (JGR, 1977). Based on these data the surgical procedure has been standardized with en bloc resection of tumor, distal and proximal normal colon, mesocolon along with apical vessels of feeding artery.

stage*	recurrence rate	5 year OS		
		colon	rectum	total
I	3.70%	90.60%	89.30%	90.60%
II	12.50%	83.60%	76.40%	81.20%
IIIa	24.10%	76.10%	64.70%	71.40%
IIIb	40.80%	62.10%	47.10%	56%

Japanese Society for cancer of colon and rectum, 1991-1996

Table 5. Recurrence rate of each stage of colorectal cancer after curative resection

A large difference has been found in the recurrence rate and prognosis in stage II and III colorectal cancer according to the report of 2004 in Japan. (Table 5.) (Japanese Society for Cancer of the Colon and Rectum, [JSCCR] Guidelines 2010 for the Treatment of Colorectal Cancer. (In Japanese). Similar data has also been published from other countries (Andre et



al., 2009). Only in stage III colon cancer does the prognosis depend upon the number of lymph node metastasis.

Lymph node (LN) involvement is an important prognostic indicator of carcinomas arising in the colon and the rectum. It also influences treatment decisions, as patients with node-positive colorectal carcinoma (CRC) are generally advised for systemic adjuvant chemotherapy. Thus, the accuracy of number of lymph node metastasis becomes a very important factor. However, when the precision of diagnosis of metastasis to lymph nodes is concerned factors such as length of dissection of bowel, lymph node dissection techniques and treatment of the specimen needs to be considered. In addition, the retrieval method may also depend on the facility, institutional protocol or the individual surgeon.

If the degree of LNM is inadequately assessed this changes the stage of the disease which in turn reflects on the further inappropriate treatment protocol and prognosis. Then the question of stage migration arises. However, inappropriate retrieval of lymph node and thus incorrect staging cannot be blamed on stage migration. Rectal cancer was excluded from this study as most of our patients did not undergo neoadjuvant chemo radiotherapy as patients in the west but underwent pelvic lymphadenectomy which increased the number of lymph nodes retrieved that may alter the surgical procedure preoperative stage and thus the prognosis.

There have been several papers regarding relationship between degree of LNM and its prognosis like the number of LNM, the site of metastatic lymph nodes, number of lymph nodes retrieved, and lymph node ratio (LNR). Our data showed colon cancer with only 1 LNM had significantly better prognosis than 2 or more LNM and patients with 7 or more LNM had the worst prognosis. Our recategorization considered here with both number and level is similar to the LN category in the 7th edition of TNM classification.

There are some reports that the prognosis of colon cancer is associated with number of LNM (Vaccaro et al., 2004) or classification by the number of LNM predicts prognosis better than classification by level of LNM (Carlos et al., 2004). On the other hand, Newland et al showed the level of LNM rather than the number of LNM is the most important variable associated with prognosis. (Newland et al., 1994) Tapper and Nelson et al. mentioned that staging for colorectal cancer required retrieval of 12-17 lymph nodes. (Tapper et al., 2001; Nelson et al., 2001) Kim et al. mentioned that retrieval of >10 lymph nodes offered almost certain identification of metastasis to lymph nodes, and tumor differentiation and T stage seemed to correlate with higher nodal metastasis rate. (Kim et al., 2006)

The 7th ed. TNM classification mentions that histological examination of a regional lymphadenectomy specimen will ordinarily include 12 or more lymph nodes. The most accepted limit for accurate staging seems to be at least 12 nodes, as also suggested by other current node metastasis related publications (Wittekend et al., 2003; Greene et al., 2002). Cserni suggested not only the minimum number of LNs should be considered in terms of staging but some qualitative features may also influence the accurate staging. The question arises whether accurate staging can be reached with fewer than 12 LNs or not (Cserni et al., 1999). However, the number of lymph nodes obtained in specimens of colorectal cancer is significantly associated with the length of resected bowel, patient age and tumor location (Shen et al., 2009). Recent study by Cserni et al have mentioned that nodal status of CRCs may be adequately assessed by examining the lymph nodes from the close fraction around the tumor and the 3 cm side long bowel segment in both directions (Cserni et al., 2011). Thus, the retrievable number of lymph nodes depends on different factors like stage, technique of dissection and the treatment of specimen.

In recent years, it has been reported that lymph node ratio (LNR) i.e., the number of tumor infiltrated nodes divided by the total number of resected nodes, is associated with prognosis (Schumacher et al., 2007; Vaccaro et al., 2009). LNR is a more accurate prognostic parameter than just the presence of lymph nodes metastasis (Rosenberg et al., 2008). However, this idea needs more verification as LNR changes according to the extracted lymph nodes number. The size of the LNs is one such possible qualifier in the study about diagnosis for lymph node metastasis. Cserni et al found that the evaluation of the seven largest LNs gives a correct qualitative (negative vs positive) nodal status in 97% of the cases. (Cserni G, 2002). Thus, the diagnosis and staging the degree of lymph node metastasis seems to be still controversial.

## 5. Conclusion

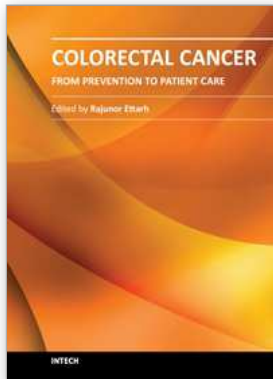
LNM staging reflects the prognosis of the disease. The method of evaluation of LNM varies according to surgical treatment, the handling of specimens, pre and post operative chemo-radiotherapy protocols etc. which varies between different institutions and countries. Therefore, it is difficult to compare every data in detail. Since, staging systems are based on depth of invasion and lymph node metastasis which influences the management and prognosis of the disease thus a standard protocol reflecting the best method for dissection of nodes and handling of specimens is necessary which reflects the accurate stage and thus the prognosis of the disease. Further studies for lymph node staging are thus necessary to find a universally accepted technique and staging system with maximum validity and reliability.

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## **Colorectal Cancer - From Prevention to Patient Care**

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The projections for future growth in the number of new patients with colorectal cancer in most parts of the world remain unfavorable. When we consider the substantial morbidity and mortality that accompanies the disease, the acute need for improvements and better solutions in patient care becomes evident. This volume, organized in five sections, represents a synopsis of the significant efforts from scientists, clinicians and investigators towards finding improvements in different patient care aspects including nutrition, diagnostic approaches, treatment strategies with the addition of some novel therapeutic approaches, and prevention. For scientists involved in investigations that explore fundamental cellular events in colorectal cancer, this volume provides a framework for translational integration of cell biological and clinical information. Clinicians as well as other healthcare professionals involved in patient management for colorectal cancer will find this volume useful.

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