THE APPLICATION VALUE OF NANO-CARBON TRACER IN LAPAROSCOPIC RADICAL RESECTION OF COLORECTAL CANCER

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ABSTRACT

AIM: To explore the application value of nano-carbon tracer in laparoscopic radical resection of colorectal cancer in the number of lymph nodes, micro-lymph nodes and lymph node metastasis.

Methods: A retrospective analysis of 66 colorectal cancer patients who met the inclusion criteria in the Gastrointestinal Surgery Department of Jingzhou First People's Hospital from July 2020 to July 2021 was retrospectively analyzed. There were 32 cases in the experimental group and 34 cases in the control group. The diameter and number of lymph nodes detected after the two groups were recorded, and the number of lymph nodes with a diameter of 5 mm or less was counted, and all lymph nodes were stained with conventional HE. Perform statistical analysis on all results.

Results: The two groups of patients were not statistically significant in gender, age, tumor differentiation, tumor invasion depth, tumor location, intraoperative blood loss (P>0.05), and statistically significant in operation time (P<0.05). A total of 597 lymph nodes were removed in the experimental group and 534 lymph nodes were detected in the control group. The average number of detections in the experimental group [(18.66±3.23) pieces] was higher than that of the control group [(15.71±3.22) pieces], and the difference was statistically significant (P<0.05). The number of lymph nodes in all patients in the experimental group was ≥12 pieces, while 21.43% of patients in the control group had <12 lymph nodes, the difference was statistically significant (P<0.05). The number of lymph nodes ≤ 5 mm in the experimental group was 371, accounting for 62.14% of the total lymph nodes, and the number of lymph nodes ≤ 5 mm in the control group was 296, accounting for 55.43% of the total lymph nodes. There was a statistically significant difference between the two groups in the number of micro-lymph nodes (diameter≤5mm) detected (P<0.05). The number of positive lymph nodes detected in the experimental group was 38 pieces (6.80%) and 24 pieces (4.71%) in the control group. There was no statistical significance in detecting lymph node metastasis (P=0.515). In the experimental group, 12 patients (35.29%) were diagnosed as stage III by pathology, and 9 patients (26.47%) in the control

group were diagnosed as stage III. There was no statistical significance in the number of patients diagnosed as stage III (P=0.430).

Conclusions: In laparoscopic radical resection of colorectal cancer, the preoperative endoscopic injection of nano-carbon tracer can locate the tumor, and guide the doctor during the operation to remove the lymph nodes and increase the number of lymph nodes detected. It is helpful to improve the accuracy of postoperative pathological staging and formulate individualized adjuvant treatment after surgery.

Keywords: colorectal cancer; laparoscopy; nano carbon tracer; number of lymph nodes; tiny lymph nodes; HE staining; lymph node metastasis;

INTRODUCTION

Colorectal Cancer (CRC) is one of the most common malignant tumors in the digestive system. The GLOBOCAN study of the International Agency for Research on Cancer (IARC) shows that there will be 1880725 new cases of colorectal cancer worldwide in 2020, Accounting for 9.8% of the overall cancer incidence rate, ranking third, with 915,880 deaths, accounting for 9.2% of the overall cancer mortality rate, ranking second (1). At present, surgical resection is still the main treatment for colorectal cancer surgery (2). In the past 20 years, with the rapid development of laparoscopic high-definition displays, 3D laparoscopy, automatic pneumoperitoneum, and various intracavitary instruments, laparoscopic colorectal cancer surgery has gradually become the mainstream technology in colorectal cancer surgery (3). Many factors affect the prognosis of patients with colorectal cancer, such as the accuracy of pathological TNM staging, R0 resection, postoperative complications, postoperative adjuvant treatment, etc. Among them, pathological TNM staging is a very important prognostic factor (4). The 8th edition of the AJCC guidelines pointed out that the number of lymph nodes seized during surgery is related to the survival rate of patients after surgery. Patients with colorectal cancer still need to seize at least 12 lymph nodes to get accurate pathological staging when undergoing radical surgery (5). Lymph node tracers tend to lymph nodes. The use of lymph node tracers during tumor surgery can better assist the surgeon in locating the tumor and clearing the surrounding lymph node tissue. As a third-generation lymph node tracer, nano-carbon has the advantages of fewer adverse reactions, clear and obvious visual field, and longer dyeing visual field compared with the first-generation tracer methylene blue and the second-generation tracer activated carbon (6). At present, nano-carbon has been demonstrated in the radical resection of thyroid cancer, breast cancer, lung cancer, gastric cancer, colorectal cancer, pancreatic cancer, and cervical cancer in the treatment of lymph nodes (7-12). To explore the application value of nano-carbon tracer in laparoscopic radical resection of colorectal cancer in the number of lymph nodes, micro-lymph nodes, and lymph node metastasis.

MATERIALS AND METHOD

Normal Information:

A retrospective analysis of case-control trials was used to collect 66 patients with colorectal cancer who met the inclusion criteria in the Gastrointestinal Surgery Department of Jingzhou First People's Hospital from July 2020 to July 2021, including 32 cases in the experimental group and 34 cases in the control group. The inclusion criteria are: (1) Preoperative colonoscopy biopsy was diagnosed as colorectal malignant tumor; (2) No liver or lung metastasis before operation; (3) No history of abdominal surgery before; (4) No history of radiotherapy and chemotherapy before operation; (5) The surgical methods were all patients undergoing laparoscopic radical resection of colorectal cancer; (6) All patients were completed by the same surgical team. Exclusion criteria: (1) previous history of other tumors; (2) patients with incomplete relevant data; (3) patients with major heart and cerebrovascular diseases; (4) patients with emergency surgery due to intestinal obstruction and perforation. This study has been approved by the Ethics Committee of Jingzhou First People's Hospital.

Experimental and Method:

Nano carbon injection:

In the experimental group, the nano-carbon suspension was injected under colonoscopy one day before the operation. A total of 1ml of nano-carbon suspension was injected with the colonoscopy needle at 3-4 points at 0.5-2cm around the edge of the tumor, and the colonoscopy injection needle was tilted. After entering the intestinal mucosa and drawing bloodless, inject about 0.25-0.35ml nano-carbon suspension per point. After the injection, slowly withdraw the needle to avoid the black-staining of the surrounding tissues after the nano-carbon injection. Routine surgical treatment is performed on the day of surgery; In the control group, titanium clip positioning was routinely performed one day before operation, and surgical treatment was routinely performed on the day of operation; 66 cases of laparoscopic colorectal cancer radical resection were performed by the same group of surgeons.

Surgery and lymph node detection:

All patients were operated in accordance with the principles of complete mesocolic excision (CME) or total mesorectal excision (TME). After the specimens were cut, they were immersed in formalin, and the number of lymph nodes was tested by the surgeon and pathologist. The diameter and number of lymph nodes detected in the experimental group and the control group were recorded respectively, and the number of lymph nodes with a diameter ≤ 5mm was counted, and all the detected results were analyzed.

HE staining:

After numbering the lymph nodes of each patient in the experimental group and the control group, after embedding in paraffin, two consecutive sections (4μm thick) were performed, and routine HE staining was performed. Tumor cells seen under the HE staining microscope are called positive.

Statistical Analysis:

Using SPSS 23.0 statistical software, measurement data is expressed as (x±s), independent sample t test is used, count data is χ2 test, P<0.05 indicates that the difference is statistically significant.

RESULTS AND DISCUSSION

Results:

Analysis and comparison of two groups of general data:

There were 66 patients with colorectal cancer who met the inclusion criteria. There are 32 patients in the experimental group, including 22 male patients and 10 female patients; aged 31-79 years old, with an average of (57.72±11.19) years old. Tumor location: 6 cases of right colon cancer, 10 cases of left colon cancer, 16 cases of rectal cancer; Tumor differentiation degree: 7 cases of poorly differentiated adenocarcinoma, 8 cases of moderately differentiated adenocarcinoma, 17 cases of highly differentiated adenocarcinoma; depth of invasion: 4 cases of mucosal layer, 5 cases of muscular layer, 23 cases of serous layer. There are 34 patients in the control group, including 18 male patients and 16 female patients; age 28-79 years old, average (56.94±12.53) years old; Tumor location: 9 cases of right colon cancer, 9 cases of left colon cancer, 16 cases of rectal cancer; tumor differentiation: 5 cases of poorly differentiated adenocarcinoma, 11 cases of moderately differentiated adenocarcinoma, 18 cases of well differentiated adenocarcinoma; Infiltration depth: 2 cases of mucosal layer, 10 cases of muscular layer, 22 cases of serous layer. There was no statistically significant difference between the two groups of patients in terms of gender, age, tumor differentiation, tumor invasion depth, tumor location, and intraoperative blood loss (P>0.05), and there was statistical significance in the length of operation (P<0.05), see Table 1.

	Control group	test group	t/χ^2	P
Number of cases (n, piece)	34	32		
age（χ±s, year）	56.94±12.53	57.72±11.19	0.265	0.792
gender				
male	18	22	1.726	0.189
Female	16	10		
Degree of differentiation				
Poorly	5	7		
Medium	11	8	0.776	0.679
Highly	18	17		
Depth of invasion				

Mucosal layer	2	4		
Muscle layer	10	5	2.297	0.317
Serosal layer	22	23		
Tumor site				
Right half	9	6		
Left half	9	10	0.593	0.744
rectum	16	16		
Operation time（min）	262.03±60.99	228.75±54.47	2.341	0.022
Intraoperative blood loss（ml）	84.41±52.41	80.00±59.51	0.319	0.751

Table 1: Comparison of general information of the two groups of patients

Comparison of the number and diameter of lymph nodes found after operation between the two groups:

A total of 597 lymph nodes were detected in the experimental group and 534 in the control group. The average number of detections in the experimental group [(18.66±3.23) pieces] was higher than that of the control group [(15.71±3.22) pieces], the difference was statistically significant (P<0.05). The number of lymph nodes in all patients in the experimental group was ≥12 pieces, while 21.43% of patients in the control group had <12 lymph nodes, the difference was statistically significant (P<0.05). The number of lymph nodes ≤5mm in the experimental group was 371, accounting for 62.14% of the total number of lymph nodes, and the number of lymph nodes ≤5mm in the control group was 296, accounting for 55.43% of the total number of lymph nodes. In terms of detection of the number of small lymph nodes (diameter <5mm) The difference between the two groups was statistically significant (P<0.05), see Table 2.

	Control group	test group	t/χ^2	P
Number of cases（n, pieces）	34	32		
Total number of lymph nodes (n, piecess)	534	597		
Average number of lymph nodes (χ±s, pieces)	15.71±3.22	18.66±3.23	3.71	0.001
Number of lymph nodes ≥ 12	28	32		
Number of lymph nodes <12 number (%)	6（21.43）	0		*Fisher*=0.025

Number of lymph node diameter <5mm (%)	296（55.43）	371（62.14）		
Average number of micro-lymph nodes (χ±s, pieces)	8.71±2.28	11.59±2.31	5.11	0.001

Table 2: Comparison of the number and diameter of lymph nodes between the two groups

Comparison of conventional pathological HE staining in the detection of lymph node metastasis and postoperative pathological staging between the two groups:

The number of positive lymph nodes detected in the experimental group was 38 pieces (6.80%) and 24 pieces (4.71%) in the control group. The results were not statistically significant (P=0.515). Twelve patients in the experimental group were diagnosed as stage III by pathology, and 9 patients in the control group were diagnosed as stage III. The results were not statistically significant (P=0.430). However, 35.29% of the people in the experimental group were diagnosed as stage III higher than 26.47% in the control group.

Postoperative complications and adverse drug reactions in the two groups:

Both groups of patients successfully completed the operation, and all followed the principles of complete mesocolic excision (CME) or total mesorectal excision (TME). There was no intraoperative hemorrhage and conversion to laparotomy in the two groups. After the operation, 1 case of abdominal cavity infection occurred in the experimental group and the control group, and there was no intestinal obstruction or intestinal fistula in the two groups. There were no obvious adverse reactions in the experimental group after the injection of nano-carbon, but there were 8 patients with nano-carbon intraperitoneal overflow during the operation.

Discuss:

Although laparoscopic surgery for rectal cancer has made significant progress, some patients with early rectal cancer still die of cancer recurrence after surgery. The reason may be the insufficient number of intraoperative lymph node dissections and the presence of lymph node micrometastasis (13-15). Studies by Bilimoria and Nathan et al. have shown that more than 48% of colorectal cancer patients have less than 12 lymph nodes, which is related to individual differences in patients, surgeons, pathologists, and other factors (16, 17). A case study by Kuijpers et al. showed that pathologists are more likely to ignore tiny lymph nodes with a diameter of 5 mm or less when collecting lymph nodes, and the proportion of colorectal cancer patients who are suitable for adjuvant chemotherapy due to insufficient lymph node sampling has reached 17.3% (18). Destri and other studies have shown that when the number of lymph nodes in stage II patients is less than 12, the postoperative disease-free survival rate (Disease-Free Survival, DFS) is significantly reduced (14). A meta-analysis on colon cancer by Chang et al. showed that increasing the number of lymph nodes can increase the survival rate of patients with stage II and III colon cancer (19). Therefore, it is very important for the prognosis of patients with colorectal cancer whether the lymphatic dissection during the operation is thorough.

Many studies have shown that the use of effective lymph node tracers in surgery can increase the number of intraoperative lymph node dissections and the number of small lymph nodes seized (20). Laparoscopic surgery lacks tactile guidance, making lymph node dissection more difficult. Compared with the traditional way of visually observing and touching and picking up lymph nodes, the use of lymph node tracers can improve the contrast between lymph nodes and surrounding tissues, making it easier for the surgeon to distinguish between lymph nodes and surrounding tissues with naked eyes, and it can also effectively avoid damage to blood vessels and surrounding tissues. Postoperative pathologists can also detect lymph nodes by observing the color changes of lymph nodes, thereby reducing the burden on pathologists. Nano-carbon is the third-generation lymph node tracer, which has the advantages of fewer adverse reactions, a clear visual field, and a longer dyeing visual field.

In this study, the operation time of the two groups in the experimental group was significantly lower than that in the control group [(228.75±54.47) min vs. (262.03±60.99) min, P<0.05], but there was no significant difference in the amount of intraoperative blood loss. (P>0.05). In terms of the number and diameter of lymph nodes seized, 597 lymph nodes were seized in the experimental group and 534 lymph nodes were seized in the control group. The average number of lymph nodes in the experimental group was significantly higher than that in the control group [(18.66±3.229) pieces vs. (15.71±3.224) pieces, P<0.05]. In terms of the number of detected micro lymph nodes, the average number of micro lymph nodes in the experimental group was significantly higher than the average number of micro lymph nodes in the control group [(11.59±2.31) pieces vs. (8.71±2.28) pieces, P<0.05]. The number of lymph nodes detected in all patients in the experimental group was ≥12 pieces, while 21.43% of the patients in the control group had less than 12 lymph nodes, which indicates that nano-carbon has a good tracing effect and can detect more lymph nodes. It also shows that the reason for the seizure of more lymph nodes may be related to the seizure of more tiny lymph nodes, and the number of lymph nodes in the experimental group is more in line with the AJCC and NCCN guidelines.

Wang and Tang's research also showed that the application of nano-carbon tracers to laparoscopic radical resection of colorectal cancer can increase the number of lymph nodes seized, but it has no significant significance in the comparison of lymph node metastasis, which is similar to our research results (21, 22). Although this study clearly shows that the use of nano-carbon tracer in laparoscopic radical colorectal resection can effectively reduce the operation time and seize more lymph nodes, it is not statistically significant in terms of lymph node metastasis and pathological staging. (P=0.515, P=0.430). But in terms of percentages, the positive percentage of lymph nodes detected in the experimental group was 6.80% higher than that in the control group, 4.71%, and the number of stage III patients in the experimental group was 35.29% higher than the control group's 26.47%. The possible reasons are 1. It is affected by the subjective factors of the surgeon or pathologist; 2. The sample size of this study is too small and the performance is not obvious; 3. Nanocarbon has a lymph node tendency, but it does not affect whether it is a positive lymph node. Conclusion; 4. For metastatic lymph nodes, the tumor tissue may oppress or block the lymphatic vessels, resulting in impaired lymphatic drainage and affecting the visualization of the lymph nodes. In the experimental group of this study, there were

8 patients with nano-carbon extravasation into the abdominal cavity, which caused the abdominal cavity to be contaminated. Although it did not affect the surgical method and safety, in the end, it still had a certain impact on the intraoperative anatomy. Therefore, the application effect of nano-carbon in colorectal cancer still needs a large number of prospective randomized controlled studies to further demonstrate.

Although there are many experiments that prove the good effect of nanocarbon on tumor localization and lymph node imaging in gastrointestinal malignant tumors, there are few reports of related adverse reactions. However, there is no clear guide on how to use nanocarbon in surgery. For how to use nano-carbon to label tumors and lymph nodes, it is necessary to make guidelines, such as nano-carbon injection time, injection site, injection method, injection dose, etc., all require a large amount of data research to prove. In this study, there was an intra-abdominal overflow of nano-carbon, but it did not affect the safety of the operation, but it increased the difficulty of the operation. In the future long-term process, whether the overflow of nano-carbon will cause aggravation of abdominal adhesion and tumor implantability Hazards such as transfer and long-term retention in the body are unforeseeable. This needs our attention.

CONCLUSION

In laparoscopic radical resection of colorectal cancer, the preoperative endoscopic injection of nano-carbon tracer can locate the tumor, and guide the doctor during the operation to remove the lymph nodes and increase the number of lymph nodes detected. It is helpful to improve the accuracy of postoperative pathological staging and formulate individualized adjuvant treatment after surgery.

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