Prognosis and influencing factors of patients with different lymph node statuses after pathological complete response to neoadjuvant chemoradiotherapy in rectal cancer

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Abstract. Neoadjuvant chemoradiotherapy (nCRT) has been shown to improve outcomes for patients with rectal cancer, but the impact of the lymph node status after achieving pathological complete response remains elusive. The present study aimed to assess the prognosis and influencing factors of patients with rectal cancer with different lymph node statuses after achieving a pathological complete response to nCRT. The clinical data of 203 patients enrolled from Hengshui People's Hospital and Hebei Cangzhou Hospital of Integrated Traditional Chinese and Western Medicine from January 2010 to December 2020 were retrospectively analyzed. These patients had undergone preoperative nCRT and were pathologically classified as 'tumor stage 0 and lymph node status 0 after neoadjuvant therapy' (ypT0N0) or 'tumor stage 0 and residual tumor in regional lymph nodes after neoadjuvant therapy' (ypT0N+) postoperatively. After surgery, patients were followed up to evaluate tumor recurrence, metastasis and survival, including disease-free survival (DFS) and overall survival (OS). Cox proportional hazards models were used to analyze factors affecting DFS and OS. Among the 203 patients included, there were 127 cases in the ypT0N0 group and 76 cases in the ypT0N+ group. The median follow-up time for the entire cohort was 56 months (range, 13-107 months). Furthermore, 72 patients (35.5%) experienced recurrence, including 53 cases (26.1%) of distant metastasis, 10 cases (4.9%) of local recurrence and 9 cases (4.4%) of both distant metastasis and local recurrence. The 5-year OS and DFS rates for all patients were 82.3 and 77.3%, respectively. The 5-year OS rates for the ypT0N0 and ypT0N+ groups were 96.1 and 59.2% (P<0.0001), respectively, and the 5-year DFS rates were 88.2 and 59.2% (P=0.0002), respectively. In addition, clinical tumor stage (cT)3-4, clinical lymph node stage (cN)+, elevated serum carcinoembryonic antigen (CEA) and postoperative pathology (ypT0N+) were independent risk factors affecting OS and DFS. In summary, the results of the present study indicate that patients with ypT0N0 rectal cancer can achieve a good long-term prognosis after nCRT. Notably, postoperative treatment and follow-up should be performed for patients with ypT0N+ and those with elevated pre-nCRT CEA, cT3-4 and cN+ stages.

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Abbreviations: pCR, pathological complete response; nCRT, neoadjuvant chemoradiotherapy; DFS, disease-free survival; OS, overall survival; CEA, carcinoembryonic antigen

Key words: rectal cancer, nCRT, pCR, influencing factors, prognosis

Introduction

Rectal cancer is a common malignant tumor of the digestive system with an increasing incidence rate in China. According to the National Cancer Center's 2024 statistics, colorectal cancer incidence and mortality rates have been rising in recent years. In 2022, there were 517,100 new cases of colorectal cancer in China, with an incidence rate of 20.1 per 100,000 individuals. Males accounted for 307,700 cases, while females for 209,400 cases, with a higher incidence observed in males. Additionally, there were 240,000 deaths, corresponding to a mortality rate of 8.56 per 100,000. The incidence of colorectal cancer increases with age, particularly among those aged 40 and above. Urban areas show higher incidence rates compared to rural areas, with the South of China exhibiting the highest rates. Notably, the proportion of low rectal cancer is relatively high in China, accounting for ~60-75% of all rectal cancer cases (1). Low rectal cancer accounts for 60-70% of all rectal cancer cases (2). Due to the insidious onset of rectal cancer, patients are often diagnosed at a locally advanced stage and/or with distant metastasis. Currently, neoadjuvant chemoradiotherapy (nCRT), surgical treatment and postoperative adjuvant therapy are standard modalities for

treating mid-low locally advanced rectal cancer (3,4). nCRT works to reduce tumor volume and achieve downstaging, thereby increasing the resection rate, sphincter preservation rate, reducing local recurrence rate, as well as prolonging disease-free survival (DFS) and overall survival (OS) (5). A total of 10-30% of patients achieve a pathological complete response (pCR) after nCRT (6,7). Compared with patients with non-pCR, patients with pCR have notably lower local recurrence and distant metastasis rates, and prolonged DFS (8,9). However, the impact of lymph node status after neoadjuvant therapy (ypN) on the prognosis of patients with 'tumor stage 0 after neoadjuvant therapy' (ypT0) still remains unclear. For instance, the incidence of lymph node metastasis has been reported to be 6-20% in patients with pCR after nCRT, which is an important factor affecting postoperative local recurrence and distant metastasis (10,11). In recent years, the treatment effect of rectal cancer has markedly improved with the continuous development of neoadjuvant treatment strategies. For instance, in the EORTC 22921 study, 13.7% of patients receiving neoadjuvant chemoradiotherapy (nCRT) achieved a pathological complete response (pT0), compared to 5.3% in the radiotherapy-only group (P<0.0001). Additionally, nCRT significantly reduced the tumor size and the number of involved lymph nodes, and the local recurrence rate was 7.6% in the nCRT group, compared to 17.1% in the radiotherapy-only group (12). Furthermore, the study by Braendengen *et al* (13) showed a 5-year overall survival rate of 70% in the nCRT group, significantly higher than the 60% in the radiotherapy-only group, with a corresponding decrease in local recurrence. However, despite regression of the primary tumor, certain patients still have lymph node metastasis, which adversely affects their long-term survival (14).

With the advancement of molecular biology techniques, increasingly more studies have begun to assess the molecular characteristics of rectal cancer and their impact on treatment response (15). Specific gene mutations, epigenetic modifications and microenvironmental factors may all affect the efficacy of nCRT and the pCR rate (16-18). These studies provide new ideas for individualized treatment and suggest that multiple factors need to be considered comprehensively during treatment to achieve the best therapeutic effect. Accordingly, to assess the relationship between different ypN statuses and prognosis, the present study retrospectively analyzed the clinical data of 203 patients with mid-low rectal cancer who achieved ypCR after nCRT. Through in-depth analysis of these clinical data, the results may provide valuable references for clinical practice and further reveal the key factors affecting the prognosis of rectal cancer.

Materials and methods

Study recruitment and design. The present study had a retrospective case-control design. Based on the inclusion and exclusion criteria, the clinical data of 203 patients who received preoperative nCRT and were postoperatively pathologically classified as ypT0N0 and ypT0N+ at Hebei Cangzhou Hospital of Integrated Traditional Chinese and Western Medicine (Cangzhou, China) from January 2010 to December 2020, were analyzed. The electronic medical records and clinical databases of the two participating institutions were accessed and reviewed from June 2021 to December 2023 to extract and verify the relevant patient data. Final follow-up was completed on January 1, 2024. The second participating institution is Hengshui People's Hospital (Hengshui, China).

The inclusion criteria before nCRT were as follows: i) Tumor lower edge within 10 cm from the anal verge, with adenocarcinoma confirmed by biopsy; ii) clinical staging as clinical tumor stage (cT)3-4 or clinical lymph node stage (cN)+; iii) no distant metastasis; and iv) no previous systemic treatment (chemotherapy, immunotherapy or radiotherapy). The inclusion criteria after nCRT were as follows: i) Treatment via radical surgery; ii) pathologically classified as ypT0 regardless of ypN classification; and iii) >3 years of follow-up after nCRT. The exclusion criteria were as follows: i) Diagnosis of inflammatory bowel disease or hereditary colorectal cancer, including familial adenomatous polyposis or Lynch syndrome; ii) diagnosis of synchronous unresectable multiple primary cancers; and iii) R1 resection performed.

The present study complied with the relevant provisions of the Helsinki Declaration and was approved by the Ethics Committee of Hebei Cangzhou Hospital of Integrated Traditional Chinese and Western Medicine (approval no. 2021-KY-062.1). All patients and/or their families provided written informed consent to participate in the study.

nCRT and postoperative adjuvant chemotherapy. All patients received intensity-modulated radiotherapy at 45.0-50.4 Gy, 1.8-2.0 Gy per session, for a total of 25-28 sessions. The neoadjuvant chemotherapy regimen was composed of concurrent single-agent capecitabine (1,000 mg/m^2 orally, twice daily and 5 days a week for 5 weeks) during radiotherapy. Surgery was performed 8-13 weeks after nCRT following the principles of total mesorectal excision. Postoperative adjuvant chemotherapy regimens included the XELOX regimen (oxaliplatin, 130 mg/m^2 intravenously on day 1; and capecitabine, 1,000 mg/m^2 orally twice daily on days 1-14, every 3 weeks) and the FOLFOX regimen (oxaliplatin, 85 mg/m^2 intravenously on day 1; leucovorin, 400 mg/m^2 intravenously on day 1; and fluorouracil, 400 mg/m^2 intravenously on day 1, then 1,200 mg/m^2/day for 2 days as a continuous intravenous infusion). The decision-making of postoperative adjuvant chemotherapy was performed jointly by surgeons and oncologists based on the pathological results of the patients.

Preoperative assessment and evaluation of nCRT efficacy. Pre- and post-nCRT cT and cN stages were evaluated using pelvic MRI. The circumferential proportion of the tumor occupying the intestinal lumen was comprehensively determined by colonoscopy, pelvic MRI and digital rectal examination. All postoperative pathological specimens from gastrointestinal tumors were reviewed by two senior pathologists to determine the final pathological results. The tumor regression grading system was used as an indicator to evaluate nCRT efficacy (19). In addition, ypT0 was defined as the complete disappearance of tumor cells under the microscope in the tumor specimen resected after nCRT (20).

Follow-up. Data related to the follow-up of the enrolled patients were collected from inpatient medical records, as well as through outpatient reviews and regular telephone visits. Starting from the end of surgery, outpatient follow-up was performed every

Table I. Comparison of general clinical data between the ypT0N0 and ypT0N+ groups.

Characteristic	ypT0N0 group (n=127)	ypT0N+ group (n=76)	χ^2 value	P-value
Age, years			0.016	0.900
<60	49 (38.6)	30 (39.5)		
≥60	78 (61.4)	46 (60.5)		
Sex			0.666	0.415
Female	66 (52.0)	35 (46.1)		
Male	61 (48.0)	41 (53.9)		
Distance to anal verge, cm			0.040	0.842
≤5	67 (52.8)	39 (51.3)		
>5	60 (47.2)	37 (48.7)		
Proportion of circle[a], %			0.040	0.841
≤50	65 (51.2)	40 (52.6)		
>50	62 (48.8)	36 (47.4)		
Degree of tumor differentiation			0.106	0.948
Low	53 (41.7)	30 (39.5)		
Intermediate	64 (50.4)	40 (52.6)		
High	10 (7.9)	6 (7.9)		
cT stage			0.031	0.960
cT1-2	32 (25.2)	20 (26.3)		
cT3-4	95 (74.8)	56 (73.7)		
cN stage			1.962	0.161
cN0	40 (31.5)	17 (22.4)		
cN+	87 (68.5)	59 (77.6)		
CEA			3.605	0.058
Normal	59 (46.5)	25 (32.9)		
Elevated	68 (53.5)	51 (67.1)		
CA 19-9			0.179	0.672
Normal	48 (37.8)	31 (40.8)		
Elevated	79 (62.2)	45 (59.2)		

[a]Proportion of circle refers to the extent of the tumor's circumference involved in the intestinal wall, as assessed through imaging techniques such as MRI or CT. Data are presented as n (%). ypT0N0, tumor stage 0 and lymph node status 0 after neoadjuvant therapy; ypT0N+, tumor stage 0 and residual tumor in regional lymph nodes after neoadjuvant therapy; cT, clinical tumor; cN, clinical lymph node; CEA, carcinoembryonic antigen; CA 19-9, carbohydrate antigen 19-9.

3 months for the first 3 years, every 6 months until 5 years postoperatively, and annually after 5 years. The follow-up ended on January 1, 2024. Follow-up included routine digital rectal examination, complete blood count, liver and kidney function tests, tumor markers, chest CT, liver ultrasound or enhanced MRI, as well as colonoscopy. OS was defined as the time from surgery to death or the last follow-up. DFS was calculated as the time to the last follow-up without recurrence or metastasis.

Statistical analysis. Data analysis was performed using SPSS 27.0 (IBM Corp.). Survival analysis was performed using the Kaplan-Meier method and the log-rank test. Postoperative serum carcinoembryonic antigen (CEA) levels were assessed at the first two routine follow-up visits after surgery. Patients were stratified into 'persistently elevated' and 'normalized/decreased' CEA groups based on whether CEA was >5.0 ng/ml on both measurements. The χ^2 test was used to compare categorical variables between groups. Cox regression was used for univariate and multivariate analysis of factors affecting OS and DFS. A two-sided $P<0.05$ was considered to indicate a statistically significant difference. To minimize time truncation bias due to variable follow-up durations, all survival analyses were right-censored at a fixed cutoff date of January 1, 2024. Additionally, 'surgery year' was extracted from the electronic medical record based on the date of radical resection following nCRT, and modeled as a continuous covariate (ranging from 2010-2020) in the univariate Cox regression analysis. This variable was used to evaluate whether patients treated in earlier years, (who had longer potential follow-up periods) exhibited different survival outcomes, and to assess whether time-related bias may have affected the observed results. The resulting hazard ratio (HR) values reflect the change in risk per 1-year

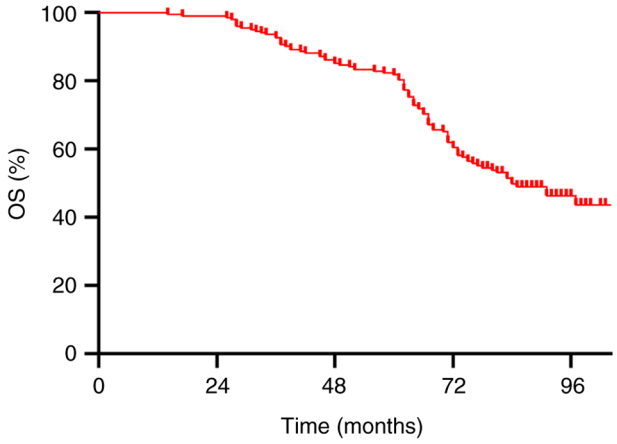

Figure 1. 5-year OS rate of the 203 patients in the present study. OS, overall survival.

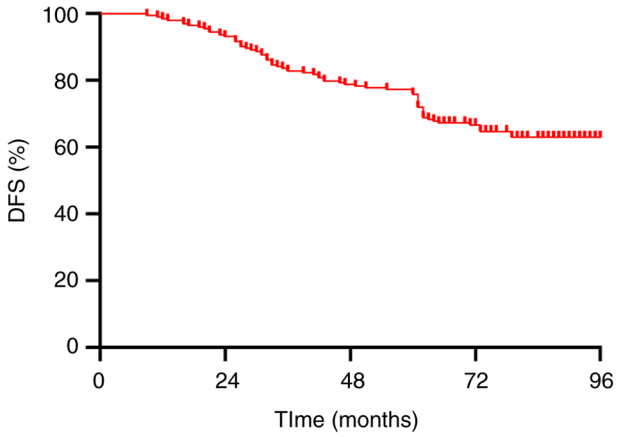

Figure 2. 5-year DFS rate of the 203 patients in the present study. DFS, disease-free survival.

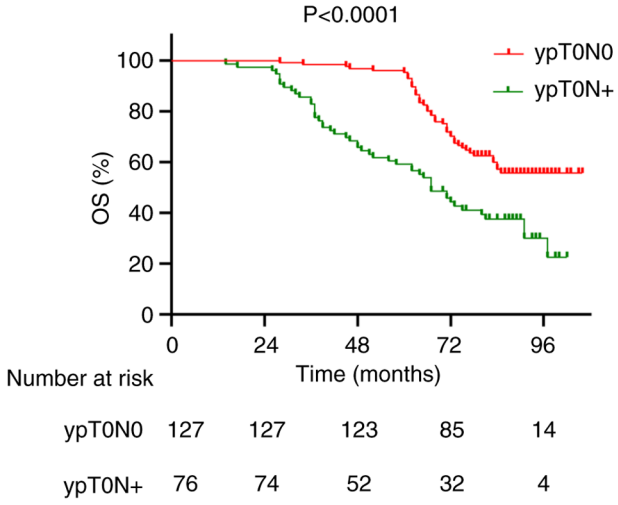

Figure 3. Comparison of the 5-year OS rate between the ypT0N0 and ypT0N+ groups. OS, overall survival; ypT0N0, tumor stage 0 and lymph node status 0 after neoadjuvant therapy; ypT0N+, tumor stage 0 and residual tumor in regional lymph nodes after neoadjuvant therapy.

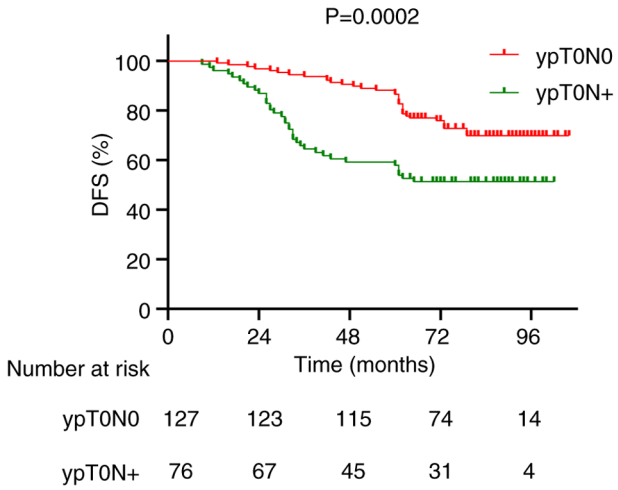

Figure 4. Comparison of the 5-year DFS rate between the ypT0N0 and ypT0N+ groups. DFS, disease-free survival; ypT0N0, tumor stage 0 and lymph node status 0 after neoadjuvant therapy; ypT0N+, tumor stage 0 and residual tumor in regional lymph nodes after neoadjuvant therapy.

increase in the surgery year (HR/year), without standardization or transformation of the variable. This covariate was included solely for the purpose of assessing time truncation bias and was not interpreted as a clinically meaningful prognostic factor due to the lack of statistical significance.

Results

Clinical characteristics analysis. Of the 203 patients included in the present study, 78 were from Hengshui People's Hospital and 125 from Hebei Cangzhou Hospital of Integrated Traditional Chinese and Western Medicine. In the Hengshui cohort, there were 42 men and 36 women, with a mean age of 60.4±9.1 years. In the Cangzhou cohort, there were 60 men and 65 women, with a mean age of 61.2±10.3 years. Furthermore, among 203 patients included in the study, there were 127 cases in the ypT0N0 group and 76 cases in the ypT0N+ group. However, there were no significant inter-group differences for any of the general clinical characteristics analyzed, such as age, sex and distance from the anal verge (all P>0.05; Table I).

Comparison of OS and DFS. The 5-year OS and DFS rates for all patients were 82.3 and 77.3%, respectively (Figs. 1 and 2). The 5-year OS rates for patients in the ypT0N0 and ypT0N+ groups were 96.1 and 59.2%, respectively (P<0.0001; Fig. 3), whilst the 5-year DFS rates were 88.2 and 59.2% for the ypT0N0 and ypT0N+ groups, respectively (P=0.0002; Fig. 4). In addition, Kaplan-Meier survival analyses were performed to assess the prognostic value of postoperative CEA levels. Patients in the 'Persistently Elevated' CEA group had significantly worse 5-year OS rates compared with those in the 'Normalized/Decreased' CEA group (64.3 vs. 86.9%; P=0.0192; Fig. 5). Similarly, the 5-year DFS rate was significantly lower in the 'Persistently Elevated' group compared with in the 'Normalized/Decreased' group (54.7 vs. 80.7%; P=0.0142; Fig. 6).

Analysis of factors influencing OS and DFS. The results of multivariate analysis demonstrated that cT3-4 before nCRT [HR=5.941; 95% confidence interval (CI), 3.036-11.625;

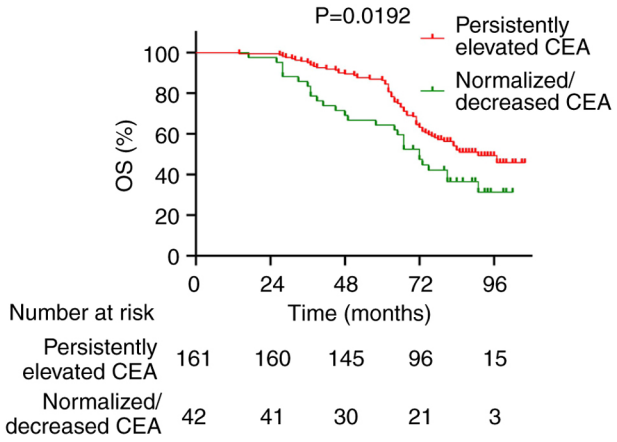

Figure 5. Kaplan-Meier survival curves for OS stratified by postoperative CEA levels: Persistently Elevated CEA vs. Normalized/Decreased CEA. OS, overall survival; CEA, carcinoembryonic antigen.

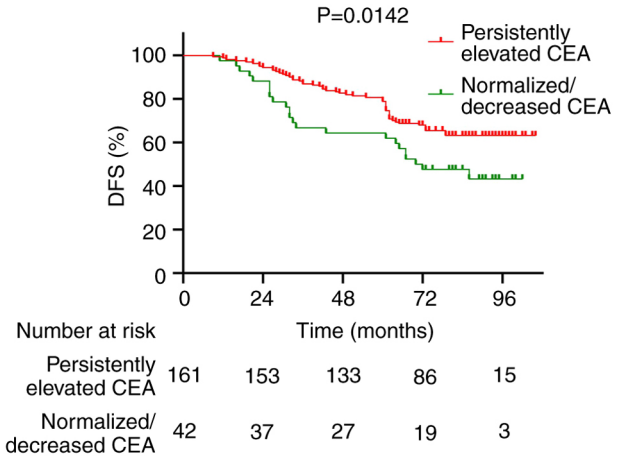

Figure 6. Kaplan-Meier survival curves for DFS stratified by postoperative CEA levels: Persistently Elevated CEA vs. Normalized/Decreased CEA. DFS, disease-free survival; CEA, carcinoembryonic antigen.

P<0.001], cN+ (HR=2.636; 95% CI, 1.576-4.411; P<0.001), elevated serum CEA level (HR=2.735; 95% CI, 1.678-4.459; P<0.001) and postoperative pathology ypT0N+ (HR=2.394; 95% CI, 1.604-3.573; P<0.001) were independent risk factors affecting OS (Table II). Moreover, the independent risk factors influencing DFS were cT3-4 (HR=4.818; 95% CI, 2.270-10.226; P<0.001), cN+ (HR=2.641; 95% CI, 1.426-4.893; P=0.002), elevated serum CEA level (HR=1.820; 95% CI, 1.079-3.071; P=0.025) and postoperative pathology ypT0N+ (HR=2.280; 95% CI, 1.426-3.644; P<0.001) (Table III).

Follow-up duration and time truncation adjustment. The follow-up duration for the entire cohort ranged from 13-107 months, with a median follow-up time of 56 months. To minimize the potential impact of time truncation bias due to variation in follow-up duration, all survival data were uniformly right-censored at a fixed cutoff date of January 1, 2024, which represented the final follow-up point for the present study. In addition, a univariate Cox regression analysis was performed including surgery year as a continuous variable to evaluate whether differences in the timing of treatment affected survival outcomes. The results demonstrated that surgery year was not significantly associated with either OS (HR=1.387; 95% CI, 0.787-2.445; P=0.258; Table II) or DFS (HR=1.006; 95% CI, 0.563-1.795; P=0.895). This suggests that time-related bias did not significantly influence survival analyses in this cohort.

Comparison of recurrence. Of the 203 patients enrolled in the present study, 72 cases (35.5%) experienced recurrence during the follow-up period, with 35 cases (27.6%) in the ypT0N0 group and 37 cases (48.7%) in the ypT0N+ group (χ^2=9.271; P=0.002). Regarding the pattern of recurrence, 53 patients (26.1%) developed distant metastasis, with 26 cases (20.5%) in the ypT0N0 group and 27 cases (35.5%) in the ypT0N+ group. There were 10 cases (26.1%) of local recurrence, with 5 cases (3.9%) in the ypT0N0 group and 5 cases (6.6%) in the ypT0N+ group. In addition, 9 patients (4.4%) experienced both distant metastasis and local recurrence, including 4 cases (3.1%) in the ypT0N0 group and 5 cases (6.6%) in the ypT0N+ group. Statistical analysis revealed no significant difference in the comparison of the pattern of recurrence between the two groups (χ^2=0.074; P=0.963) (Table IV).

Discussion

In recent years, marked progress has been made in the evaluation, staging and treatment of rectal cancer, leading to the improvement of the oncological prognosis of patients. The application of nCRT contributes further to improving local tumor control and reducing adverse drug reactions (21), particularly for patients with postoperative pCR, resulting in markedly extended DFS (22). Sell *et al* (23) performed a study on 370 patients with rectal cancer who underwent preoperative chemoradiotherapy and reported that 50 patients (13.5%) achieved pCR, with 5-year OS and DFS rates of 95 and 92%, respectively; this is similar to the results of the present study. Moreover, through the follow-up of 58 patients with mid-low locally advanced rectal cancer who achieved pCR after nCRT with a median time of >4 years, Campos-Lobato *et al* (24) reported that the distant metastasis rate in pCR patients was notably lower compared with non-pCR patients, with a 5-year OS rate of 93%. A multicenter study including 566 ypT0N0 patients from 36 centers also reported that the local recurrence rate, distant metastasis rate, 5-year DFS and OS were 1.6, 8.9, 85.0 and 90.0%, respectively, in patients after a median follow-up time of 46.4 months (25). In the present study, the 5-year DFS and OS of 127 ypT0N0 patients were 88.2 and 96.1%, respectively, similar to the aforementioned reports.

The ypN status is an important factor for the prognosis of patients with locally advanced rectal cancer after nCRT (26). Among the 203 ypT0 patients in the present study, 76 had positive lymph node remnants. Moreover, the present study demonstrated that residual mesorectal lymph node metastasis after nCRT, even after the complete regression of the primary tumor in the rectal wall, adversely affected OS and DFS. In light of these findings, the results of the present study were further compared with those of several previous publications addressing lymph node status, pCR and prognostic evaluation after nCRT. Li Destri *et al* (27) assessed the impact of nCRT on lymph node retrieval and prognosis in 142 patients with rectal cancer. Their findings highlighted the prognostic

Table II. Influencing factors of overall survival of the 203 patients in the present study.

	Univariate analysis			Multivariate analysis		
Characteristic	HR	95% CI	P-value	HR	95% CI	P-value
Age (≥60 vs. <60 years)	1.248	0.825-1.888	0.293			
Sex (female vs. male)	1.139	0.768-1.689	0.518			
Distance to anal verge (>5 vs. ≤5 cm)	1.095	0.737-1.625	0.654			
Proportion of circle[a] (≤50 vs. >50%)	1.005	0.677-1.491	0.980			
cT stage (cT3-4 vs. cT1-2)	4.024	2.091-7.745	<0.001	5.941	3.036-11.625	<0.001
cN stage (cN+ vs. cN0)	1.903	1.153-3.141	0.012	2.636	1.576-4.411	<0.001
CEA (elevated vs. normal)	3.502	2.161-5.673	<0.001	2.735	1.678-4.459	<0.001
CA 19-9 (elevated vs. normal)	1.142	0.763-1.709	0.520			
ypT0N stage (ypT0N+ vs. ypT0N0)	2.349	1.582-3.486	<0.001	2.394	1.604-3.573	<0.001
Lymph node involvement (≥3 vs. 1-2)	1.504	1.027-3.459	0.097			
Surgery year	1.387	0.787-2.445	0.258			

[a]Proportion of circle refers to the extent of the tumor's circumference involved in the intestinal wall, as assessed through imaging techniques such as MRI or CT. HR, hazard ratio; CI, confidence interval; ypT0N0, tumor stage 0 and lymph node status 0 after neoadjuvant therapy; ypT0N+, tumor stage 0 and residual tumor in regional lymph nodes after neoadjuvant therapy; cT, clinical tumor; cN, clinical lymph node; CEA, carcinoembryonic antigen; CA 19-9, carbohydrate antigen 19-9.

Table III. Influencing factors of disease-free survival of the 203 patients in the present study.

	Univariate analysis			Multivariate analysis		
Characteristic	HR	95% CI	P-value	HR	95%CI	P-value
Age (≥60 vs. <60 years)	1.487	0.900-2.456	0.121			
Sex (female vs. male)	1.193	0.751-1.895	0.455			
Distance to anal verge (>5 vs. ≤5 cm)	1.053	0.663-1.673	0.826			
Proportion of circle[a] (≤50 vs. >50%)	1.162	0.732-1.845	0.525			
cT stage (cT3-4 vs. cT1-2)	3.367	1.613-7.027	0.001	4.818	2.270-10.226	<0.001
cN stage (cN+ vs. cN0)	2.017	1.106-3.679	0.022	2.641	1.426-4.893	0.002
CEA (elevated vs. normal)	2.301	1.373-3.856	0.002	1.820	1.079-3.071	0.025
CA 19-9 (elevated vs. normal)	1.087	0.672-1.758	0.735			
ypT0N stage (ypT0N+ vs. ypT0N0)	2.322	1.461-3.690	<0.001	2.280	1.426-3.644	<0.001
Lymph node involvement (≥3 vs. 1-2)	1.391	0.957-3.018	0.219			
Surgery year	1.006	0.563-1.795	0.895			

[a]Proportion of circle refers to the extent of the tumor's circumference involved in the intestinal wall, as assessed through imaging techniques such as MRI or CT. HR, hazard ratio; CI, confidence interval; ypT0N0, tumor stage 0 and lymph node status 0 after neoadjuvant therapy; ypT0N+, tumor stage 0 and residual tumor in regional lymph nodes after neoadjuvant therapy; cT, clinical tumor; cN, clinical lymph node; CEA, carcinoembryonic antigen; CA 19-9, carbohydrate antigen 19-9.

value of the lymph node ratio (LNR) and the potential staging inaccuracy due to reduced lymph node yield post-nCRT. By contrast, the present study did not focus on lymph node count or LNR, but instead identified ypT0N+ patients as a distinct high-risk subgroup based on pathological nodal involvement despite complete tumor regression. This offers more direct implications for postoperative treatment planning. Furthermore, Zhang *et al* (28) evaluated 432 patients with rectal cancer and identified predictors for achieving pCR, such as low baseline CEA and extended interval between nCRT and surgery. Their results support a 'watch-and-wait' strategy for ypT0N0 patients. However, the present study specifically excluded ypT0N0 patients and focused on ypT0N+ individuals who exhibited inferior survival outcomes, thereby underscoring the necessity of intensified adjuvant therapy in this subgroup. Zhu *et al* (29) analyzed 482 patients and reported that the total number of examined lymph nodes markedly affected staging accuracy and long-term survival. Whilst this emphasizes the importance of adequate lymph node dissection, the results of the present study indicate that even patients

Table IV. Recurrence pattern comparison between ypT0N0 and ypT0N+ groups.

Parameter	ypT0N0 group (n=127)	ypT0N+ group (n=76)	χ^2 value	P-value
Recurrence cases	35 (27.6)	37 (48.7)	9.271	0.002
Distant metastasis	26 (20.5)	27 (35.5)	0.016	0.899
Local recurrence	5 (3.9)	5 (6.6)	0.009	0.925
Both distant and local recurrence	4 (3.1)	5 (6.6)	0.074	0.963

Data are presented as n (%). ypT0N0, tumor stage 0 and lymph node status 0 after neoadjuvant therapy; ypT0N+, tumor stage 0 and residual tumor in regional lymph nodes after neoadjuvant therapy.

with complete tumor regression can harbor nodal metastasis, which independently predicts poor prognosis. Thus, the findings of the present study shift the focus from lymph node quantity to the biological impact of ypT0N+ status. Moreover, Ozturk *et al* (30) proposed a lymph node regression grading (LNRG) system based on histopathological response in 469 patients. Although they reported that LNRG may provide additional prognostic information, the present study adopted a clinically translational approach by directly identifying ypT0N+ patients as requiring different postoperative management. Nodal regression grade was analyzed; however, survival data and risk stratification specific to the ypT0N+ population were provided. Collectively, these comparisons underscore the novelty of the present study in defining ypT0N+ as a distinct prognostic entity. Unlike previous studies that emphasize lymph node count, regression grade or pCR prediction, the present work focused on a specific, understudied subgroup and proposes actionable treatment strategies to improve their outcomes. Lu *et al* (31) analyzed 59 ypT0N0 patients after nCRT and reported that 8 patients (13.6%) had mesorectal lymph node metastasis, and the 5-year DFS and OS rates of these patients were markedly longer than those of ypT0N+ patients. Zhang *et al* (32) analyzed 76 ypT0 patients, with 9 (11.8%) classed as ypN+. They reported that the 5-year DFS and OS of ypT0N+ patients were 62.5 and 72.9%, respectively. Furthermore, multivariate analysis in the present study revealed that ypT0N+ was an independent risk factor for DFS.

In addition, the findings of the present study demonstrated that there was a lower distant metastasis rate of patients with pCR after nCRT. This may be due to the fact that during the period from the end of radiotherapy to surgery, patients who achieve pCR had no residual tumor cells, whilst patients who did not achieve pCR may have had a continuous interaction between tumor cells and the surrounding environment, markedly increasing the possibility of distant metastasis. Therefore, a good response of the primary tumor to nCRT can be regarded as a sign of systemic treatment effectiveness. However, unlike the tumor in the rectal wall, lymph node metastasis did not completely disappear with the achievement of pCR in the rectal wall tumor. This may be attributed to the continuous evolution of tumor cells in primary and metastatic sites, further enhancing their invasive, metastatic and chemoradiotherapy-resistant malignant potentials (33,34). Therefore, even if the tumor in the rectal wall disappears completely, metastatic cells in the lymph nodes can survive and lead to distant metastasis that produces an adverse impact on patient prognosis.

In the present study, independent risk factors affecting OS and DFS included cT stage, cN stage and serum CEA levels. A study performed in the United States, which included 23,747 patients with rectal cancer, reported that the rate of pCR gradually decreased as the cT stage (cT1-cT4) and cN stage (cN0-cN2) changed, with a lower rate of pCR in patients with a higher stage and cN+ compared to those with patients a lower stage and cN0 (P<0.001), indicating a worse prognosis (35). Peng *et al* (36) also reported that cT stage (P=0.043) and N stage (P=0.003) could predict pCR rates. Patients with a larger tumor burden often experience resistance to radiotherapy. Considering the different growth rates of blood vessels and tumors, patients with a larger tumor burden tend to have a larger proportion of hypoxic tumor cells, and the hypoxic microenvironment may promote tumor progression, increase tumor heterogeneity, and enhance tumor tolerance to radiotherapy and chemotherapy (37). Zhou *et al* (38) analyzed 124 patients with locally advanced rectal cancer and reported that pre-treatment elevation of CEA was an independent high-risk factor affecting OS in ypT0N patients. Colloca *et al* (39) performed a meta-analysis on 20 studies and demonstrated that patients in the elevated CEA group had markedly prolonged 3- and 5-year DFS rates compared with the normal pre-treatment CEA group. CEA has been reported to affect the biological behavior of tumor cells through autocrine secretion, increasing tumor cell survival, inhibiting tumor cell differentiation, as well as promoting endothelial cell activation and tumor angiogenesis through paracrine secretion (40). The present study further demonstrated the prognostic value of pre-treatment CEA for patients with locally advanced rectal cancer. In addition to pre-treatment levels, the present study revealed that persistently elevated postoperative CEA was significantly associated with worse OS and DFS. This highlights the importance of serial postoperative CEA monitoring, not only for recurrence detection, but also for postoperative risk stratification and treatment planning. These findings support the integration of dynamic CEA assessment into follow-up strategies, especially for identifying patients who may benefit from intensified adjuvant therapy or closer surveillance.

For patients with elevated CEA levels, previous studies have suggested several potential improvements in treatment strategies. Firstly, personalized treatment plans are crucial for these patients. Specifically, more aggressive preoperative and postoperative treatment regimens can be considered for patients with elevated CEA. For example, enhancing the nCRT

regimen (such as introducing more potent chemotherapy agents like FOLFOX or adding irinotecan) can increase the pCR rate and improve long-term survival (41). Secondly, dynamic monitoring of CEA levels can serve as an important indicator of treatment response. Post-nCRT changes in CEA after nCRT may suggest the sensitivity to treatment in patients with elevated preoperative CEA levels. If postoperative CEA remains elevated, particularly when the local tumor has completely disappeared, it may suggest the presence of micro-residual tumors or micrometastases, requiring closer follow-up and further treatment for such patients. Therefore, regular monitoring of CEA levels is essential for predicting postoperative recurrence or metastasis in the management of patients with elevated CEA (42).

Moreover, molecular mechanisms serve a critical role in treatment response after nCRT in patients with rectal cancer. In particular, certain gene mutations and epigenetic changes can enhance tumor cell resistance to chemotherapy and radiotherapy, thereby worsening prognosis through multiple molecular pathways (43). KRAS mutations have been reported to be common in rectal cancer, especially in locally advanced cases. Although nCRT can notably reduce tumor size, the effectiveness of chemotherapy and radiotherapy is often suboptimal in KRAS-mutant patients (44). This may be attributed to the continuous activation of cell proliferation and growth signals caused by the mutations, leading to treatment resistance. NRAS and BRAF mutations are also considered important factors affecting the efficacy of nCRT, with BRAF-mutant patients generally exhibiting worse survival and a higher recurrence risk compared to BRAF wild-type patients (45). In addition, epigenetic changes such as DNA methylation and histone modifications can also have an impact on the treatment response by affecting the proliferation, migration and invasiveness of tumor cells. For instance, dysregulated DNA methylation of tumor suppressor genes is considered a mechanism contributing to tumor cell resistance to treatment. Specifically, methylation of tumor suppressor genes (such as MutL protein homolog 1 and adenomatous polyposis coli) can induce microsatellite instability (MSI), an epigenetic alteration that is associated with variable sensitivity to nCRT, with MSI-high (MSI-H) tumors often demonstrating enhanced responsiveness to immunotherapy but inconsistent responses to chemoradiotherapy. Epigenetic modifications can also alter the immune escape mechanisms and angiogenesis responses in the tumor microenvironment (TME), thus affecting the sensitivity to radiotherapy and chemotherapy (46).

Furthermore, the role of the TME in cancer progression, immune evasion and drug resistance has received increasing attention in recent decades. The TME consists of several cell types (including immune cells, fibroblasts, and endothelial cells), as well as their secreted cytokines and metabolic products. These factors can modulate the treatment response of patients and long-term survival by altering tumor progression and therapeutic resistance (47). Future research should further investigate the specific mechanisms by which the TME impacts prognosis and develop personalized treatment strategies tailored for ypT0N+ patients.

The results of the present study indicated that ypT0N+ patients had a worse prognosis compared to ypT0N0 patients, with the requirement for more aggressive treatment strategies. Therefore, based on domestic and international guidelines, and the latest research (48,49), the following specific treatment strategies are proposed: First, intensified postoperative adjuvant chemotherapy should be considered for ypT0N+ patients. Standard oxaliplatin-based regimens such as XELOX (capecitabine plus oxaliplatin) and FOLFOX (fluorouracil plus oxaliplatin) remain the recommended postoperative therapies (50). In the present study, ypT0N+ patients exhibited significantly higher recurrence rates and worse long-term survival compared with ypT0N0 patients, suggesting that current standard treatments may be insufficient in this high-risk subgroup. Although irinotecan-containing regimens such as FOLFIRI have demonstrated efficacy in the management of metastatic colorectal cancer (51), their role in the postoperative treatment of ypT0N+ patients remains unproven. These approaches are therefore considered hypothesis-generating and warrant further prospective evaluation. Second, although anti-EGFR agents (such as cetuximab) and anti-VEGF agents (such as bevacizumab) have demonstrated efficacy in metastatic colorectal cancer (52), their role in the adjuvant treatment of ypT0N+ rectal cancer remains to be established. Current clinical guidelines do not recommend the routine use of these agents in the postoperative setting for non-metastatic disease (53). However, further research should explore whether selected high-risk patients with specific genetic mutations (such as KRAS, NRAS and BRAF) could benefit from such targeted strategies. Similarly, immune checkpoint inhibitors [such as programmed cell death 1 (PD-1)/programmed cell death-ligand 1 (PD-L1) inhibitors] have shown promising efficacy in MSI-H colorectal cancers, particularly in advanced or recurrent settings (54). Whilst their role in the adjuvant treatment of ypT0N+ patients remains investigational, the presence of MSI-H or high tumor mutation burden may serve as a biomarker for future clinical trials assessing the value of immunotherapy in this subgroup. In the present study, none of the 203 patients received targeted therapy or immunotherapy as part of their adjuvant treatment. All patients were treated according to institutional protocols at the time, which were based on standard chemotherapy regimens such as XELOX or FOLFOX. Therefore, the discussion on targeted therapy and immunotherapy in ypT0N+ patients is provided to reflect current advances in the literature and highlight potential directions for future individualized treatment strategies. Lastly, ypT0N+ patients should undergo enhanced follow-ups. Regular monitoring of CEA levels, imaging (such as MRI and CT) and colonoscopy should be performed to detect potential recurrence or metastasis at an early stage. For high-risk patients (such as those with elevated CEA or KRAS mutations), there is a need to increase the follow-up frequency, with imaging assessments potentially required every 3-6 months. Collectively, treatment strategies for ypT0N+ patients should include individualized chemotherapy regimens, potential targeted therapy and immunotherapy. Further prospective studies should facilitate the determination of the optimal combination of these treatment approaches to improve therapeutic outcomes and reduce recurrence risks. Moreover, notably in the present study, ypT0N+ patients were classified as having complete primary tumor regression (ypT0) but with concurrent lymph node metastasis. Due to potential limitations in pathological specimen sampling, the identification

of certain pCR cases may not be entirely accurate, indicating the presence of false-positive pCR cases. This is particularly relevant when microscopic residual disease is not fully reflected in pathological evaluation, potentially underestimating recurrence risks in certain patients. To minimize the impact of false-positive pCR, future research should focus on standardizing pathological evaluations, expanding the scope of tissue sampling and integrating liquid biopsy techniques to enhance the accuracy of early recurrence detection. Previous studies have reported that persistent postoperative detection of circulating tumor (ct)DNA is associated with an increased risk of recurrence in patients with colorectal cancer, including those with rectal cancer (55,56). Integrating ctDNA analysis into postoperative surveillance may help identify ypT0N+ patients who require intensified adjuvant therapy and more rigorous follow-up protocols.

The present study demonstrated that elevated preoperative CEA levels are an independent risk factor for poor prognosis in patients with rectal cancer, consistent with previous findings. Zhou *et al* (38) reported that pretreatment CEA elevation was markedly associated with reduced OS in patients with ypT0N+ rectal cancer. Similarly, Peng *et al* (36) reported that cT stage, cN stage and CEA levels were notable predictors of pCR following nCRT. However, inconsistencies still exist. For example, Zhang *et al* (32) reported no significant survival differences based on preoperative CEA levels. These discrepancies may be attributed to heterogeneity in patient populations, differences in tumor location, radiotherapy regimens (such as inclusion of short-course radiotherapy) and varying definitions of CEA thresholds. Biologically, CEA promotes tumor progression through autocrine and paracrine mechanisms, enhancing tumor cell survival, inhibiting differentiation and stimulating angiogenesis. These effects may contribute to chemoradiotherapy resistance in CEA-elevated tumors. Clinically, dynamic CEA monitoring is valuable in assessing treatment response. Patients with normalized postoperative CEA levels generally have a favorable prognosis, whereas persistently elevated postoperative CEA (even in the absence of detectable residual tumor) may indicate micro-metastatic disease or occult recurrence risk (57). In the cohort in the present study, patients with persistently elevated postoperative CEA had significantly worse 5-year OS (64.3 vs. 86.9%; P=0.0192) and DFS rates (54.7 vs. 80.7%; P=0.0142) compared with those whose CEA normalized after surgery. Based on this, CEA-based postoperative risk stratification is proposed: Patients with normalized postoperative CEA may follow standard surveillance intervals, and patients with persistently elevated CEA should undergo intensified follow-up and be considered for escalated adjuvant therapy. The findings of the present study align with much of the existing literature (58,59) and emphasize the importance of individualized follow-up strategies based on dynamic tumor markers (42). However, future studies are warranted to further validate the integration of CEA kinetics into treatment algorithms for patients with rectal cancer.

Micro (mi)RNAs serve a direct role in rectal cancer progression and resistance to nCRT, as demonstrated in studies assessing their impact on chemoradiotherapy outcomes. For example, De Palma *et al* (60) reported that miR-21 enhances resistance to nCRT by inhibiting apoptosis-related pathways and upregulating tumor cell survival signals. Conversely, Li *et al* (47) reported that lower expression of miR-223 was associated with an improved response to chemoradiotherapy. These findings suggest that miRNA profiling could serve as a biomarker for predicting nCRT efficacy in patients with rectal cancer.

KRAS mutations are common in rectal cancer and are closely associated with the development of lymph node metastasis (61). Studies have reported that KRAS mutations not only affect the biological behavior of the primary tumor, but are also associated with an increased risk of lymph node metastasis (43). In ypT0N+ patients, KRAS mutations could contribute to the development of lymph node metastasis. Treatment strategies targeting KRAS mutations, such as anti-EGFR therapy (for example, cetuximab), may improve therapeutic outcomes, particularly in patients with lymph node metastasis after neoadjuvant therapy. Prognostically, KRAS mutations are associated with a worse outcome, warranting more aggressive postoperative treatment and close follow-up (62).

BRAF mutations, particularly the V600E mutation, drive tumor progression in rectal cancer through constitutive activation of the MAPK/ERK signaling pathway, promoting cell proliferation, angiogenesis and resistance to apoptosis (63). These mutations are associated with poor prognosis, higher recurrence rates and increased lymph node involvement (43). In patients with rectal cancer, BRAF mutations have been associated with lower pCR rates after nCRT and inferior survival outcomes (64). Although BRAF-targeted therapies have shown efficacy in metastatic colorectal cancer, particularly when combined with MEK inhibitors and anti-EGFR agents (65), there is currently limited evidence supporting their use in patients with locally advanced rectal cancer with residual nodal disease after nCRT. Further studies are warranted to investigate whether such strategies may be beneficial for the ypT0N+ subgroup.

MSI is caused by mutations or deletions in DNA mismatch repair genes, resulting in genetic instability (66). MSI-H is commonly found in rectal cancer and is associated with lower rates of lymph node metastasis and improved responses to chemotherapy (67). However, certain MSI-H patients may still exhibit ypT0N+ status, possibly due to immune evasion mechanisms within the tumor (68). For these patients, immune checkpoint inhibitors (such as PD-1 inhibitors) could be considered as part of their treatment plan. Moreover, immunotherapy has been reported to be highly effective in MSI-H colorectal cancers, particularly in cases of recurrence or metastasis (69).

Furthermore, the present study evaluated the prognostic value of the number of positive lymph nodes. Although the univariate Cox regression analysis did not show a statistically significant association between lymph node count (1-2 vs. ≥3) and OS or DFS (both P>0.05), this may be due to the limited sample size in the ≥3 positive node subgroup (n=21), which may have reduced the power to detect a difference. Nonetheless, lymph node burden remains a clinically relevant consideration. Prior studies have reported that patients with higher numbers of positive nodes are at greater risk for recurrence and worse long-term outcomes (27,70-72). Therefore, lymph node count should still be incorporated into postoperative risk stratification, especially in ypT0N+ patients, to guide treatment intensity and follow-up frequency.

However, the present study has several limitations. Whilst no confirmed false-positive pCR cases were identified in the present study, the potential for sampling bias and microscopic residual disease must be acknowledged. The classification of ypT0N+ patients as having achieved pCR in the primary tumor was based on standard histopathological examination, which may not always capture residual tumor cells hidden within fibrotic tissue. Additionally, the absence of liquid biopsy or molecular residual disease assessment limited the ability to verify complete tumor eradication. Potential false-positive pCR cases could have led to an underestimation of recurrence risk and an overestimation of nCRT effectiveness, reinforcing the need for intensified postoperative surveillance in ypT0N+ patients. Future studies should incorporate advanced molecular and imaging techniques, including ctDNA analysis, to improve the accuracy of pCR assessment and guide treatment decisions. Furthermore, in the present study, treatment response categories were defined as follows: Complete response was defined as ypT0N0, indicating no residual tumor at both the primary and nodal sites; partial response included ypT1-2N0 or ypT0N+ cases, where tumor regression was observed; stable disease included ypT3-4N0 or persistent positive nodes without significant regression; and progressive disease was characterized by evidence of disease progression or distant metastasis. These classifications align with standard rectal cancer response criteria and were used to assess treatment outcomes in our cohort. However, due to the retrospective design of the present study and the lack of routine molecular testing protocols at the study institutions during the study period (2010-2020), genetic and immunological data such as KRAS, NRAS, BRAF mutations, MSI and PD-1/PD-L1 expression were not available for analysis. These biomarkers are critical determinants of prognosis and treatment efficacy in patients with rectal cancer, particularly for identifying patients who might benefit from targeted therapies or immunotherapy. The absence of these molecular data prevented the performance of a more precise risk stratification and tailored therapeutic recommendations for ypT0N+ patients. Future studies, ideally prospective in design, should incorporate comprehensive genetic and molecular profiling to further elucidate their prognostic significance and improve individualized treatment strategies. Additionally, due to the retrospective design, the present study relied on existing medical records, which limited the availability of certain prognostic variables, including lifestyle factors such as diet, body mass index, smoking, alcohol consumption and physical activity. These factors have been associated with rectal cancer outcomes in previous studies (73-75), but their impact could not be assessed in the present study due to a lack of systematic documentation. Additionally, whilst patients were enrolled from two institutions, the study remains regionally specific and single-center in nature, which may limit the generalizability of the findings. Variations in chemotherapy regimens and radiotherapy doses were also present, although their effect on survival was not statistically significant. Furthermore, the relatively short follow-up period for certain patients could influence survival estimates. Therefore, future research should include prospective lifestyle data collection, longer follow-up periods and multi-center validation to further refine prognostic models and treatment strategies for patients with rectal cancer. Lastly, standard Cox proportional hazards models were employed to assess survival outcomes without incorporating time-dependent covariates. Although the proportional hazards assumption was satisfied, time-dependent covariates were not incorporated into the final multivariate analysis, which represents a methodological limitation. To minimize the potential impact of time truncation bias due to variable follow-up durations, a uniform right-censoring strategy was applied by setting January 1, 2024 as the fixed cutoff date for all survival analyses. Additionally, surgery year was included as a continuous variable in the univariate Cox regression model, with no significant association with OS or DFS, suggesting minimal confounding from differences in enrollment time. Future studies should consider using extended Cox models or longitudinal statistical methods to better account for dynamic variables such as postoperative biomarker changes.

In summary, patients with locally advanced rectal cancer who achieve ypT0N0 after preoperative nCRT have a good prognosis, whereas those with ypT0N+ have a worse prognosis. Therefore, in future studies, the definition of pCR should be limited to ypT0N0, and postoperative chemotherapy strategies for patients with ypT0N+ may need to be adjusted accordingly. Meanwhile, ypN+, pre-nCRT elevated CEA levels, cT3-4 and cN+ are independent risk factors for OS and DFS in ypT0 patients, emphasizing the need for enhanced postoperative treatment and follow-up for these patients.

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Availability of data and materials

The data generated in the present study may be requested from the corresponding author.

Authors' contributions

JZ performed the data analysis and paper writing. YNZ was responsible for the research design and guided the revision of the paper. YS provided clinical cases, participated in data analysis and interpretation, and revised the manuscript critically for important intellectual content. LX contributed to the research design, provided data collection support and assisted in drafting the manuscript. HL participated in data analysis and contributed to the revision of the manuscript. XY assisted in data analysis and provided clinical case data. YK assisted in data collection and analysis, and contributed to manuscript writing. YYZ made substantial contributions to the acquisition and analysis of clinical data. WG assisted in data collection and initial analysis. JZ and YNZ confirm the authenticity of all the raw data. All authors have read and approved the final version and agreed to be accountable for the integrity of the work.

Ethics approval and consent to participate

The current study was performed in accordance with the Declaration of Helsinki and approved by the local Ethics Committee of Cangzhou Hospital of Integrated Traditional Chinese Medicine and Western Medicine (Cangzhou, China; approval no. 2021-KY-062.1). All patients and/or their families signed informed consent forms.

Patient consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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