100820_Crosscheck.docx

WORD COUNT 2861
TIME SUBMITTED 04-FEB-2025 09:59PM
PAPER ID 114414449

Retrospective analysis of preoperative tumor marker levels in rectal cancer patients: Implications for diagnosis

Abstract

BACKGROUND

Early detection of rectal cancer poses significant challenges. Current diagnostic methods, including colonoscopy, imaging techniques, and fecal tests, have limitations such as invasiveness, cost, and varying sensitivity. This study evaluated the diagnostic value of preoperative serum tumor markers in rectal cancer patients.

AIM

To investigate the value of a multi-marker approach for the preoperative diagnosis of rectal cancer.

METHODS

A retrospective analysis of 250 patients diagnosed with rectal cancer between July 2022 and July 2024 was conducted. Preoperative alpha-fetoprotein levels, carcinoembryonic antigen (CEA), cancer antigen 125 (CA125), CA19-9, CA15-3, and CA72-4 were analyzed. All blood samples were collected under standardized conditions, including fasting status and proper storage methods, within two weeks before surgery. Diagnostic performance was assessed using receiver operating characteristic curve analysis. Correlations among clinicopathological features were also evaluated.

RESULTS

CEA demonstrated the highest diagnostic performance among individual tumor markers with an area under the curve (AUC) of 0.78 [95% confidence interval (CI): 0.73-0.83]. However, a combination of CEA, CA19-9, and CA72-4 showed superior performance, achieving an AUC of 0.87 (95%CI: 0.83-0.91). Significant correlations were observed between CEA levels and several clinicopathological features, including tumor stage ($P < 0.001$), lymph node involvement ($P = 0.002$), and distant metastasis ($P < 0.001$). Furthermore, in a subgroup analysis of patients diagnosed after July 2022, the integration of fecal occult blood testing with the tumor marker panel (CEA + CA19-9 +

CA72-4) significantly improved diagnostic accuracy, increasing the AUC to 0.91 (95%CI: 0.86-0.96).

CONCLUSION

A multimarker approach combining CEA, CA19-9, and CA72-4 with fecal occult blood testing enhances the preoperative assessment of patients with rectal cancer. These findings suggest potential improvements in risk stratification and management of patients with rectal cancer.

Key Words: Rectal cancer; Tumor markers; Carcinoembryonic antigen; Cancer antigen 19-9; Cancer antigen 72-4; Fecal occult blood test; Diagnosis

Li M, Yuan DH, Yang Z, Lu TX, Zhang L. Retrospective analysis of preoperative tumor marker levels in rectal cancer patients: Implications for diagnosis. *World J Gastrointest Surg* 2025; In press

Core Tip: This study highlights the effectiveness of a multi-marker approach for the preoperative diagnosis of rectal cancer. Analysis of serum levels of the tumor markers carcinoembryonic antigen, cancer antigen 19-9, and cancer antigen 72-4, along with fecal occult blood testing, demonstrated improved diagnostic accuracy, with an area under the curve of 0.91. Significant correlations between elevated carcinoembryonic antigen levels and clinicopathological features, such as tumor stage and lymph node involvement, suggest that this combined strategy could enhance the risk stratification and management of patients with rectal cancer, ultimately aiding in earlier detection and better clinical outcomes.

INTRODUCTION

Colorectal cancer (CRC) is one of the most prevalent malignancies worldwide, with rectal cancer accounting for approximately one-third of all CRC cases[1]. Despite the

advancements in diagnostic techniques and treatment modalities, rectal cancer continues to pose significant challenges in terms of early detection, accurate staging, and prognostic assessment. In this context, tumor markers have garnered considerable attention as a potential tool for improving patient management and outcomes. Tumor markers are biochemical substances cancer cells, or other cells produce in response to certain benign conditions. These markers can be found in blood, urine, or tissue samples, indicating the presence, progression, or response to cancer treatment. In the realm of rectal cancer, several tumor markers have been investigated for their diagnostic and prognostic value, including alpha-fetoprotein (AFP), carcinoembryonic antigen (CEA), cancer antigen 125 (CA125), CA19-9, CA15-3, and CA72-4.

AFP is normally produced in the liver and yolk sac of developing fetuses. Although it is primarily associated with liver and germ cell tumors, elevated levels have been reported in some gastrointestinal cancers, including rectal cancer. CEA is perhaps the most well-studied tumor marker in CRC. It is a glycoprotein involved in cell adhesion and is often elevated in various adenocarcinomas, particularly those of the gastrointestinal tract[2]. CA125 is traditionally associated with ovarian cancer but has also been investigated in other malignancies, including CRC[3]. CA19-9 is another widely studied marker initially developed for pancreatic cancer but is also elevated in other gastrointestinal malignancies[4]. CA15-3, which is primarily used for breast cancer monitoring, and CA72-4, which is associated with gastric cancer, have also shown potential utility in CRC[5].

Although these tumor markers have been studied individually in various contexts, their collective evaluation in a specific setting of rectal cancer, particularly in the preoperative phase, remains an area of active research. The limitations of current diagnostic methods highlight the need for more reliable and non-invasive biomarkers that can complement existing techniques. Preoperative assessment is crucial as it influences treatment decisions, surgical planning, and overall patient management. Moreover, the potential of these markers to provide prognostic information could significantly impact follow-up strategies and personalized treatment approaches.

Recently, there has been a growing interest in combining multiple tumor markers to improve diagnostic accuracy. This approach, often called a tumor marker panel, aims to overcome the limitations of individual markers and provide a more comprehensive assessment of the disease state. Integrating novel markers or diagnostic tests, such as fecal occult blood test (FOBT), into these panels may enhance their clinical utility.

MATERIALS AND METHODS

Study design and patient selection

This retrospective study was conducted at Linquan County People's Hospital, China tertiary care center, and included patients diagnosed with rectal cancer between July 2022 and July 2024. Patients were systematically screened from the hospital information system based on the following inclusion criteria: Histologically confirmed rectal cancer, availability of preoperative serum tumor marker data for at least four of the six markers (AFP, CEA, CA125, CA19-9, CA15-3, and CA72-4), and completion of the necessary clinical and pathological assessments. The study protocol was approved by the Institutional Review Board of Linquan County People's Hospital, and the requirement for informed consent was waived because of the retrospective nature of the study. Patients were excluded if they had a history of other malignancies, received neoadjuvant therapy before blood sample collection, had incomplete medical records defined as missing key information (such as tumor marker levels or clinicopathological data), presented with synchronous tumors, or had a history of inflammatory bowel disease.

Data collection

Patient data were extracted from the electronic medical records and encompassed a comprehensive range of information. This included demographic details, such as age, sex, and body mass index; clinical characteristics, including tumor location and clinical stage (based on the 8th edition of the American Joint Committee on cancer tumor-node-metastasis staging system); and pathological features, such as histological type,

differentiation grade, lymph node involvement, and presence of distant metastasis. Additionally, preoperative serum levels of tumor markers (AFP, CEA, CA125, CA19-9, CA15-3, and CA72-4) and FOBT results were collected. Treatment details were also recorded, including the type of surgery and adjuvant therapy. Finally, follow-up data, including the date of the last follow-up, recurrence, and survival status, were included to facilitate a comprehensive analysis.

Tumor marker analysis

Blood samples for tumor marker analysis were collected within two weeks before surgical intervention. The specific time window for sample collection was standardized to within two weeks before surgery. Additionally, all samples were collected under consistent conditions, including fasting status when required, and were processed and stored using standardized protocols. Serum tumor marker levels were measured using commercially available immunoassay kits according to the manufacturers' instructions. AFP, CEA, CA125, CA19-9, and CA15-3 levels were analyzed using a Siemens Atellica IM1600 automated immunoassay analyzer (Siemens Healthineers, Erlangen, Germany). CA72-4 levels were measured using a Roche Cobas e411 analyzer (Roche Diagnostics, Basel, Switzerland). The following cutoff values were used to define elevated levels based on the manufacturers' recommendations and institutional standards: AFP (8.1 ng/mL), CEA (5 ng/mL), CA125 (30.2 U/mL), CA19-9 (37 U/mL), CA15-3 (32.4 U/mL), and CA72-4 (6.9 U/mL). FOBT were performed using a guaiac-based test or fecal immunochemical test, with a positive result defined as ≥ 20 μg Hb/g feces.

Statistical analysis

All statistical analyses were performed using SPSS version 26.0 (IBM Corp., Armonk, NY, United States). Statistical significance was set at $P < 0.05$ for all analyses. Descriptive statistics were used to summarize the patient characteristics and tumor marker levels. Continuous variables are presented as the median and interquartile range or mean ± SD depending on the data distribution. Categorical variables are

expressed as frequencies and percentages. The diagnostic performance of individual tumor markers and their combinations was evaluated using receiver operating characteristic curve analysis. The area under the curve (AUC), sensitivity, specificity, positive predictive value, and negative predictive value were calculated. The optimal cutoff values for each marker were determined using the Youden index.

The association between tumor marker levels and clinicopathological features was assessed using the χ^2 or Fisher's exact test for categorical variables and the Mann-Whitney U test or Kruskal-Wallis test for continuous variables. A Spearman's rank correlation coefficient was used to evaluate the correlation between tumor markers. Subgroup analysis was performed on patients diagnosed after July 2022 to assess the added value of FOBT. The diagnostic performances of the tumor marker panels with and without including FOBT results were compared using the DeLong test for AUC comparison.

Ethical considerations

This study followed the Declaration of Helsinki and the Good Clinical Practice guidelines. Patient confidentiality was maintained throughout data collection and analysis. All patient data were anonymized and de-identified before analysis.

RESULTS

Patient characteristics

A total of 250 patients with rectal cancer who met the inclusion criteria were included in this study. The median age of the cohort was 62 years (range: 35-85 years), and the male-to-female ratio was 1.4:1. Table 1 summarizes the demographic and clinicopathological characteristics of the study population.

Tumor marker levels

The preoperative serum levels of the six tumor markers were analyzed in all patients. The median and proportion of patients with elevated levels of each marker are shown

in Table 2. CEA showed the highest proportion of elevated levels (48.8%), followed by CA19-9 (24.8%) and CA72-4 (20.8%).

Diagnostic performance of tumor markers

The diagnostic performances of individual tumor markers and their combinations were evaluated using receiver operating characteristic curve analysis. Table 3 presents the AUC, sensitivity, specificity, positive predictive value, and negative predictive value for each marker and selected combinations. CEA demonstrated the highest individual diagnostic performance with an AUC of 0.78 [95% confidence interval (CI): 0.73-0.83]. The combination of CEA, CA19-9, and CA72-4 showed the best overall performance with an AUC of 0.87 (95%CI: 0.83-0.91).

Correlation with clinicopathological features

The association between tumor marker levels and clinicopathological features was also analyzed. Significant correlations were observed between CEA levels and tumor stage ($P < 0.001$), lymph node involvement ($P = 0.002$), and distant metastasis ($P < 0.001$). CA19-9 levels were significantly associated with tumor differentiation ($P = 0.018$) and distant metastasis ($P = 0.007$). CA72-4 levels significantly correlated with tumor size ($P = 0.013$) and lymph node involvement ($P = 0.009$). Table 4 presents the correlations between tumor marker levels and key clinicopathological features.

Subgroup analysis: FOBT

FOBT results were available for a subgroup of patients diagnosed after July 2022 ($n = 112$). The inclusion of FOBT in the tumor marker panel (CEA + CA19-9 + CA72-4 + FOBT) significantly improved the diagnostic performance compared to the panel without FOBT (AUC: 0.91, 95%CI: 0.86-0.96 *vs* AUC: 0.87, 95%CI: 0.81-0.93, $P = 0.028$). Table 5 presents the diagnostic performance of the tumor marker panel with and without FOBT. The addition of FOBT to the tumor marker panel increased the sensitivity from 81.2% to 86.5% and specificity from 77.8% to 82.3%.

DISCUSSION

This retrospective study evaluated the diagnostic value of preoperative serum tumor markers (AFP, CEA, CA125, CA19-9, CA15-3, and CA72-4) in patients with rectal cancer. Our findings highlight the potential utility of these markers, particularly when used in combination, to improve the preoperative assessment and management of patients with rectal cancer.

Diagnostic performance of tumor markers

CEA demonstrated the highest diagnostic performance among the individual tumor markers examined, with an AUC of 0.78. This finding is consistent with previous studies that established CEA as the most widely used tumor marker in CRC[2-6]. Our results showed that CA19-9 and CA72-4 also exhibited moderate diagnostic performance in rectal cancer, with AUCs of 0.71 and 0.69, respectively. Although these markers are not routinely used for rectal cancer screening, their potential utility has been suggested in previous studies[4-6]. Our cohort's relatively low diagnostic performances of AFP, CA125, and CA15-3 suggest that these markers may have limited value as individual diagnostic tools for rectal cancer.

A key finding of our study is the improved diagnostic performance achieved by combining multiple tumor markers. The CEA, CA19-9, and CA72-4 panels demonstrated the highest AUC (0.87), significantly outperforming all the individual markers. This supports the growing body of evidence suggesting that multi-marker panels can enhance the detection and assessment of CRC. Compared to similar studies, our research utilized a broader range of tumor markers and included a larger sample size, which may account for the higher diagnostic performance observed. Additionally, including FOBT in our panel further distinguishes our study from previous studies. The synergistic effects of combining markers may be attributed to their different molecular origins and heterogeneity in tumor biology.

Correlation with clinicopathological features

Our analysis revealed significant correlations between certain tumor markers and clinicopathological features of rectal cancer. CEA levels are strongly associated with tumor stage, lymph node involvement, and distant metastasis. These findings align with previous studies reporting CEA as a valuable indicator of disease extent and metastatic potential in CRC[7]. The observed correlations between CA19-9 levels and tumor differentiation and distant metastasis suggest that this marker may provide additional information about tumor biology and behavior. Similarly, the association of CA72-4 with tumor size and lymph node involvement indicates its potential role in assessing the local tumor extent. These findings underscore the importance of considering multiple tumor markers in the preoperative evaluation of patients with rectal cancer, as they may offer complementary information on different aspects of tumor characteristics.

Integration of FOBT

A notable aspect of our study included FOBT results for patients diagnosed after July 2022. The addition of FOBT to the tumor marker panel (CEA + CA19-9 + CA72-4) significantly improved the diagnostic performance, increasing sensitivity and specificity. This finding is particularly relevant in recent efforts to enhance CRC screening strategies. While the FOBT, particularly the fecal immunochemical test, is primarily used for CRC screening in asymptomatic individuals[8], our results suggest that it may also be valuable in the diagnostic workup of suspected rectal cancer cases. The complementary nature of serum tumor markers and FOBT highlights the potential benefits of integrating multiple testing modalities to improve diagnostic accuracy.

Clinical implications

These findings have important clinical implications. Using a multi-marker panel (CEA + CA19-9 + CA72-4) in the preoperative assessment of patients with rectal cancer may improve diagnostic accuracy and provide a more comprehensive evaluation of tumor

characteristics[9]. The strong prognostic value of preoperative CEA level supports its use in risk stratification and treatment planning, suggesting that patients with elevated CEA levels may benefit from more aggressive treatment strategies and closer postoperative surveillance. Furthermore, integrating FOBT with serum tumor markers offers a promising approach for enhancing the diagnostic workup of suspected rectal cancer cases, potentially leading to earlier detection and improved outcomes[10,11]. Compared to existing diagnostic methods, this multi-marker approach can be a non-invasive, cost-effective adjunct that complements imaging and endoscopic evaluations[12]. The observed correlations between tumor markers and clinicopathological features suggest that these markers may aid in the preoperative prediction of tumor stage and metastatic potential, potentially influencing decisions regarding neoadjuvant therapy or surgical approach. Collectively, these implications underscore the potential of tumor markers to significantly enhance the management and outcomes of patients with rectal cancer[13].

Limitations and future directions

Despite its contributions, this study had several limitations that warrant further acknowledgment. The study's retrospective nature introduces potential information biases, such as selection bias and missing data, which may have affected the validity of the findings. The single-center design may also restrict generalizability to other populations or healthcare settings. Several research directions could be pursued: (1) Conducting prospective, multicenter studies to validate the diagnostic value of the proposed multi-marker panel; (2) Investigating novel tumor markers or molecular signatures to enhance rectal cancer diagnosis accuracy; (3) Assessing the cost-effectiveness of integrating multiple tumor markers and FOBT into routine clinical practice; and (4) Exploring the potential role of tumor markers in monitoring treatment response and detecting recurrence in rectal cancer patients. Future studies should implement standardized data collection protocols to mitigate information bias and consider using blinded assessments for tumor marker evaluation. These future

endeavors will address the current limitations and advance our understanding and management of rectal cancer.

CONCLUSION

This retrospective study provides evidence of the utility of a multi-marker approach for the preoperative assessment of patients with rectal cancer. The combination of CEA, CA19-9, and CA72-4 demonstrated superior diagnostic performance compared with individual markers, and the addition of FOBT further improved diagnostic accuracy. These findings suggest that a comprehensive tumor marker panel, potentially including FOBT, could enhance the preoperative evaluation, risk stratification, and management of rectal cancer patients. Future prospective studies are needed to validate these results and explore their implementation in clinical practice, aiming to improve patient outcomes in rectal cancer.

100820_Crosscheck.docx

ORIGINALITY REPORT

3%
SIMILARITY INDEX

PRIMARY SOURCES

1	www.researchsquare.com Internet	18 words — 1%
2	www.por-journal.com Internet	16 words — 1%
3	www.frontiersin.org Internet	15 words — < 1%
4	www.science.gov Internet	15 words — < 1%
5	www.slideshare.net Internet	15 words — < 1%

EXCLUDE QUOTES	ON	EXCLUDE SOURCES	< 15 WORDS
EXCLUDE BIBLIOGRAPHY	ON	EXCLUDE MATCHES	< 15 WORDS