EFFECT OF MULTIMODAL THERAPY ON THE INCIDENCE OF CIRCULATING TUMOR CELLS IN THE BLOOD OF PATIENTS WITH RECTAL CANCER

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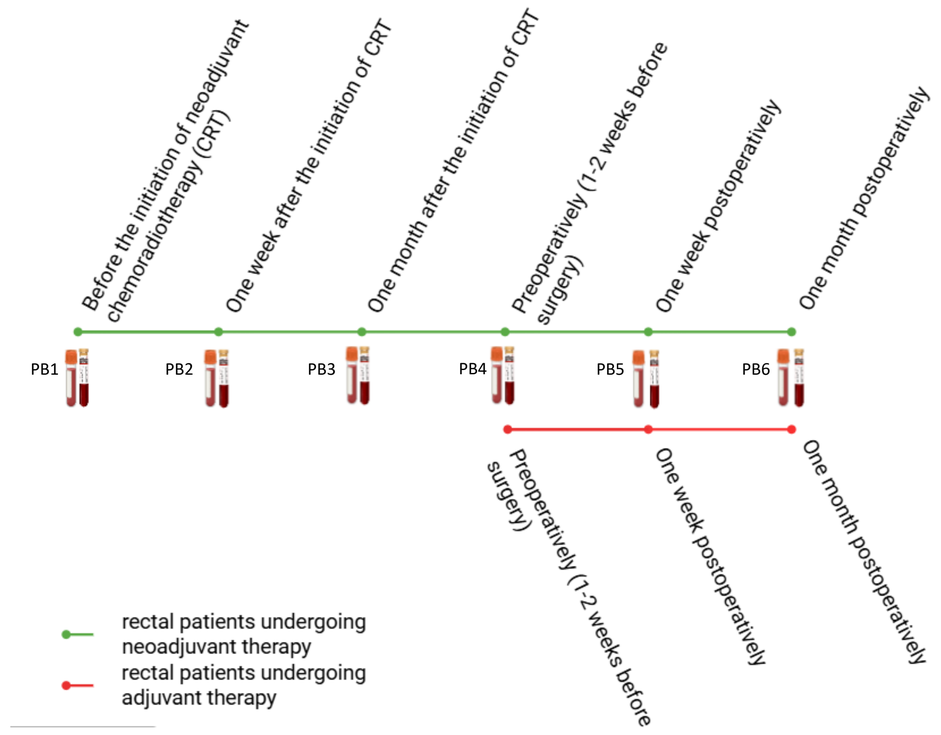
INTRODUCTION

Adjuvant and neoadjuvant therapies are essential in the treatment of rectal cancer, particularly in intermediate to advanced stages. These approaches aim to improve prognosis, reduce recurrence, and increase the chance of sphincter preservation. To date, there is no clear consensus on whether adjuvant and neoadjuvant therapies have a fundamentally different effect on the presence of circulating tumor cells (CTCs) in the peripheral blood of rectal cancer patients undergoing either adjuvant or neoadjuvant treatment.

PATIENTS

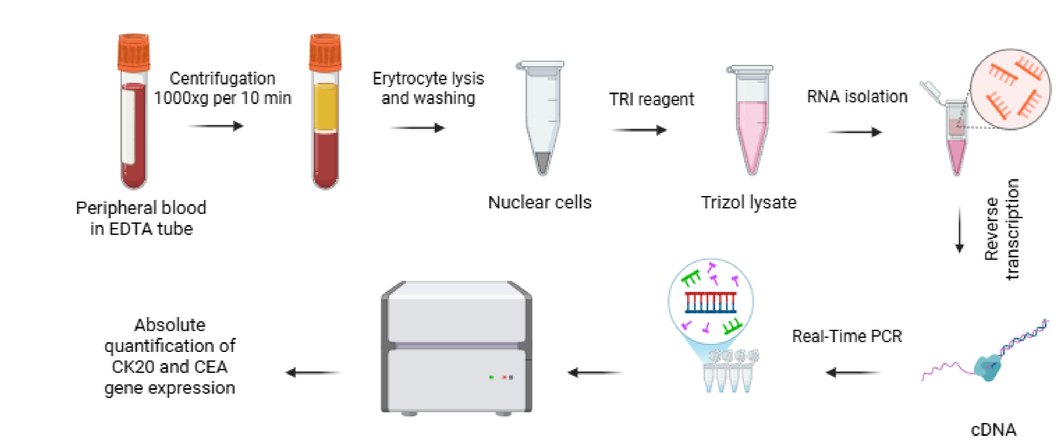
Clinical sample collection for this investigation commenced in **January 2023**. The study cohort consisted of **79 patients with histologically confirmed rectal cancer** who were enrolled according to predefined inclusion criteria. Of these, **25 were female** and **54 were male**, with a mean age of **63 years** (range, 42–80 years). Patients were stratified according to treatment modality: **20 individuals underwent neoadjuvant therapy**, while **59 received adjuvant therapy**. All participants were managed according to current clinical guidelines, and treatment allocation was based on standard oncological indications.



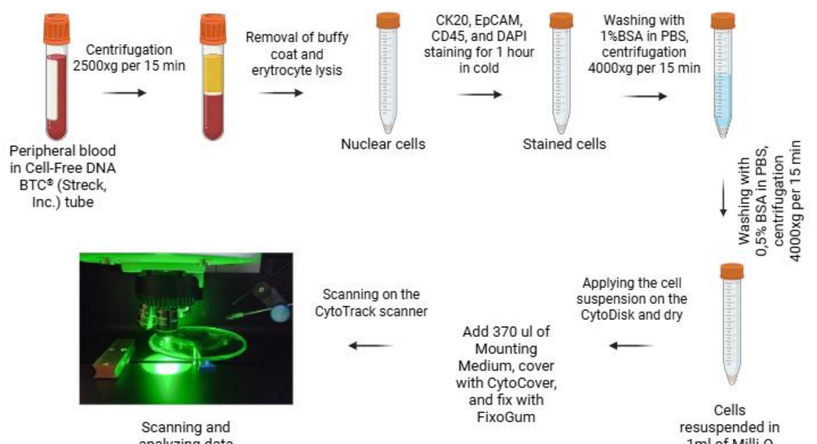
Blood samples were taken repeatedly from each patient at several time intervals depending on the type of treatment.

METHODS

Real-Time PCR analyses



CytoTrack CT11 analysis



RESULTS

Of the 79 patients, 49 had a complete set of collections. Among the 10 patients who received neoadjuvant therapy, all 6 samples were collected and analyzed. Among the 39 patients who received adjuvant chemoradiotherapy, all 3 samples were collected and analyzed.

Using Real-Time PCR detection of cytokeratin 20 gene expression, positive circulating tumor cells were detected in at least one of six collections in 9 out of 10 patients (90%) who received neoadjuvant therapy. In patients who received adjuvant therapy, CTCs were detected in at least one of three samples in 29 of 39 patients (74%).

Using CytoTrack CT11 analysis, positive CTCs were detected in at least one of six collections in 5 out of 10 patients (50%) who received neoadjuvant therapy. In patients who received adjuvant therapy, CTCs were detected in at least one of three samples in 18 of 39 patients (46%).

CTCs detection using CytoTrack C11

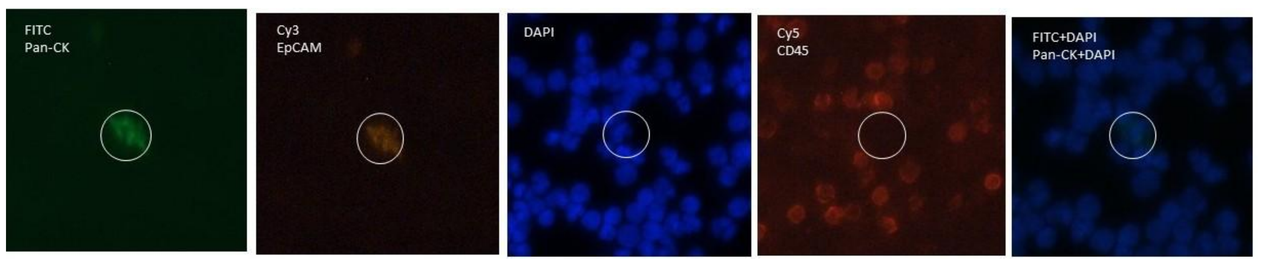


Fig.1: Detection of positive cell using CytoTrack C11 (2/C, Denmark). Pan-cytokeratin staining (FITC, green), EpCAM (Cy3, orange), DAPI (blue), CD45 (Cy5, red), and Pan-cytokeratin+DAPI (merge).

CTCs detection using Real-Time PCR detection of Cytokeratin 20 expression

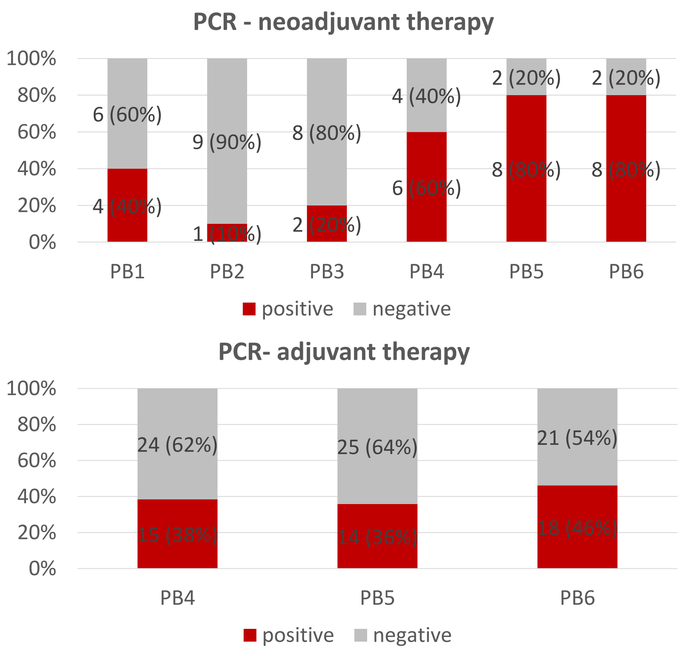


Fig. 2: Proportion of rectal cancer patients with undergoing neoadjuvant therapy according to the presence of CTCs detected by real-time PCR (CK20 gene expression). Abbreviations: PB1, PB2… = individual peripheral blood draws collected according to the study schedule.

Fig. 3: Proportion of rectal cancer patients with undergoing adjuvant therapy according to the presence of CTCs detected by real-time PCR (CK20 gene expression). Abbreviations: PB4, PB5… = individual peripheral blood draws collected according to the study schedule.

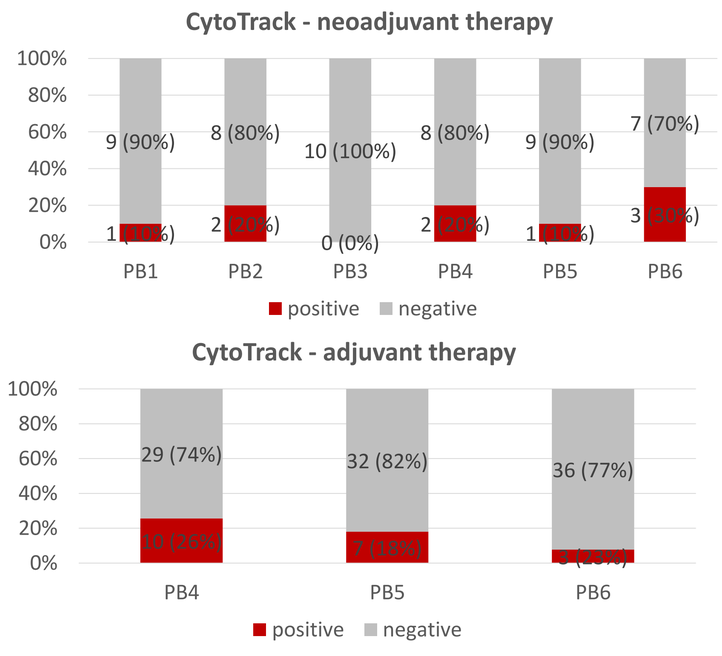


Fig. 4: Proportion of rectal cancer patients with undergoing neoadjuvant therapy according to the presence of CTCs detected by CytoTrack (pan-CK, EpCAM, DAPI positive, CD45 negative). Abbreviations: PB1, PB2… = individual peripheral blood draws collected according to the study schedule.

Fig. 5: Proportion of rectal cancer patients with undergoing adjuvant therapy according to the presence of CTCs detected by CytoTrack (pan-CK, EpCAM, DAPI positive, CD45 negative). Abbreviations: PB4, PB5… = individual peripheral blood draws collected according to the study schedule.

ACKNOWLEDGEMENT

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DISCUSSION AND CONCLUSION

The higher incidence of CTCs after neoadjuvant therapy in rectal cancer can be attributed to the biological effects of treatment (selective survival of more invasive clones, changes in the microenvironment, immunosuppression) and the technical difficulty of surgery in cases of fibrotic changes. However, elevated CK20 levels may reflect the release of cell fragments and mRNA from damaged cells due to therapy, not necessarily a higher number of viable CTCs.

Neoadjuvant therapy is typically indicated for patients with locally advanced rectal cancer or features associated with a high risk of recurrence. Monitoring CTCs may provide prognostic information, but its reliability depends on the correct choice of detection method.