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# Total Neoadjuvant Therapy Outcomes and Watch-and-Wait Feasibility in Locally Advanced Rectal Cancer: A Single-Institution Retrospective Cohort Study

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# Total Neoadjuvant Therapy Outcomes and Watch-and-Wait Feasibility in Locally Advanced Rectal Cancer: A Single-Institution Retrospective Cohort Study

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## Simple Summary

Advanced rectal cancer is increasingly treated with total neoadjuvant therapy (TNT)—a strategy delivering all chemotherapy and radiation before surgery—to improve survival and enable some patients to avoid surgery through watch-and-wait. We studied 205 patients with high-risk rectal cancer treated with a "sandwich" TNT protocol (chemotherapy before and after radiation) at our cancer center. Complete tumor disappearance occurred in 30% of patients, with excellent five-year survival (81%). Among patients choosing watch-and-wait after complete response, two-thirds preserved their rectum long-term, though careful monitoring was essential as one-third experienced tumor regrowth requiring delayed surgery. Importantly, MRI-detected high-risk features and surgical margin quality remained the strongest predictors of outcome. These findings support sandwich TNT as effective real-world treatment and confirm watch-and-wait as a viable organ-preservation option when combined with rigorous surveillance.

## Abstract

**Background/Objectives**: Total neoadjuvant therapy (TNT), integrating systemic chemotherapy and radiotherapy before surgery or surveillance, has become a standard approach for locally advanced rectal cancer (LARC). However, optimal sequencing strategies and long-term outcomes of watch-and-wait (W&W) following sandwich TNT remain insufficiently characterized. We evaluated oncologic outcomes and treatment response in patients treated with an institutional sandwich TNT protocol. **Methods**: We conducted a retrospective cohort study of consecutive patients with LARC treated with sandwich TNT (induction chemotherapy followed by hypofractionated intensity-modulated radiotherapy with simultaneous integrated boost [IMRT-SIB] chemoradiotherapy and consolidation chemotherapy) at the Institute of Oncology Ljubljana between 2016 and 2023. The primary endpoint was an overall complete response (CR; pathological [pCR] and clinical [cCR]). Secondary endpoints included tumor regression grade (TRG), major pathological response (MPR), R0 resection rate, organ preservation, overall survival (OS), and disease-free survival (DFS). **Results**: Among 205 patients (median age 61 years), overall CR was 29.5% (pCR 19.3% and cCR 10.2%). Major pathological response (TRG 3–4) occurred in 37.6%. R0 resection was achieved in 94.5%. In the W&W cohort (n=21), local regrowth occurred in 33.3% (95% CI 14.6–57.0%) over a median follow-up of 4.96 years. Surgery-free survival at 5 years was 73.1% (95% CI 55.0–97.2%). Estimated five-year OS was 81.1% (95% CI 75.5–87.2%) and 5-year DFS was 75.2% (95% CI 69.0–82.0%). In multivariable analysis, non-R0 resection (HR 6.06), MRI circumferential resection margin positivity (HR 3.11), and extramural vascular invasion positivity (HR 1.97) remained independent predictors of DFS. **Conclusions**: Sandwich TNT yields meaningful tumor response and durable survival in MRI-defined

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high-risk LARC. Structured W&W offers organ preservation with acceptable oncologic control under intensive surveillance.

**Keywords:** total neoadjuvant therapy; rectal cancer; neoadjuvant chemotherapy; radiotherapy sequencing; watch-and-wait; organ preservation; pathological complete response; circumferential resection margin; extramural vascular invasion; IMRT-SIB

## 1. Introduction

The management of locally advanced rectal cancer (LARC) has undergone continuous evolution since the late 1980s, driven by improvements in surgical technique, radiotherapy, systemic therapy, and imaging [1–6]. The introduction of total mesorectal excision (TME) by Heald and Ryall in 1986 marked a pivotal enhancement in local control, reducing local recurrence rates from the prior 30 – 40% to approximately 5 – 10% [1]. Building on TME, the Swedish Rectal Cancer Trial (1997) and the Dutch trial (2001) demonstrated that preoperative short-course radiotherapy (SCRT) further reduced local recurrence to 3 – 11% compared with surgery alone (8 – 27%), with the Swedish trial reporting a 10% absolute improvement in overall survival [2,3].

The EORTC 22921 trial (2004) and the FFCD 9203 trial (2006) demonstrated that fluorouracil added to preoperative radiotherapy halved the local recurrence rates (approximately 8% vs. 16%)[4,5]. The CAO-ARO-AIO 04 trial (2004) brought additional evidence that preoperative chemoradiotherapy (CRT) reduced local recurrence compared with postoperative CRT (6% vs. 13%, p = 0,006), with improved tolerability [6]. However, these advances in locoregional control – while highly effective with modern 5-year local recurrence rates < 5% – left unaddressed the persistent problem of distant metastatic disease, occurring in 25 – 35% of patients at 5 years [1,6]. Additionally, adjuvant chemotherapy completion remained suboptimal due to postoperative morbidity and reduced functional reserve [1,2].

From 2020 onwards, the therapeutic landscape for LARC underwent a substantial paradigm shift with the adoption of total neoadjuvant therapy (TNT) – the delivery of both systemic chemotherapy and radiotherapy entirely before surgery or surveillance [7–10]. The pivotal PRODIGE – 23 trial (2021 – 2024) randomized 461 LARC patients to induction TNT with six cycles of mFOLFIRINOX followed by CRT plus surgery and adjuvant chemotherapy, versus standard CRT followed by surgery and adjuvant chemotherapy [11,12]. With 7 – year follow – up, the induction TNT arm demonstrated significant gains: 5,1% improvement in disease – free survival (DFS), 5,8% improvement in overall survival (OS), and 6,9% improvement in metastasis – free survival. Concurrently, the RAPIDO trial (2021) demonstrated that short – course radiotherapy followed by chemotherapy before TME reduced disease – related treatment failure and approximately doubled pCR rates compared with conventional CRT [9].

Parallel advances in prognostic staging have substantially refined patient selection for TNT. The MERCURY trial (2006) established magnetic resonance imaging (MRI) as the criterion standard for staging, demonstrating 92% specificity for predicting negative circumferential resection margin (CRM) [13,14]. Subsequently, MRI – involved CRM emerged as the only preoperative staging parameter significantly correlated with OS, DFS and LR [14]. Beyond CRM, MRI – based assessment of extramural vascular invasion (EMVI), tumor deposits, and mesorectal fascia involvement guide treatment intensification [14–16]. A contemporary analysis by Lord et al. (2022) demonstrated that MRI – defined high – risk features (positive CRM, TD, or EMVI) predicted prognosis superior to TNM staging alone, underscoring the primacy of radiologic risk factors in tailoring TNT intensity [16].

Emerging evidence has validated organ – preservation strategies via watch – and – wait (W&W) in carefully selected patients achieving complete clinical response after TNT. The OPRA trial demonstrated that selective W&W after cCR/near – cCR can achieve substantial rectum preservation

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rates without apparent detriment to disease – free or overall survival, provided structured MRI and endoscopy – based surveillance with timely salvage surgery is implementation [10,17]. The International Watch – and – Wait Registry, encompassing 880 patients across multiple institutions, reported regrowth rates of approximately 20 – 25% with careful surveillance protocols; importantly, 71,4 – 100% of patients with regrowth underwent salvage total mesorectal excision with R0 resection achieved in all [18]. These data establish W&W as a feasible option for organ preservation, particularly relevant for younger patients with longer life expectancies who wish to avoid the long – term morbidity of permanent colostomy or sexual/urinary dysfunction.

Recent trials have challenged the necessity of radiotherapy in select low – risk patients. The PROSPECT trial randomized over 1,000 low – risk LARC patients (no T4, no N2, negative CRM > 3 mm) to selective radiotherapy omission in patients achieving > 20% tumor reduction after induction FOLFOX, versus standard CRT [19]. Similarly, the CONVERT trial and the FOWARC trial validated non – inferiority of chemotherapy – only approaches in carefully selected intermediate – risk disease [20,21]. Simultaneously, emerging biomarker approaches – including circulating tumor DNA (ctDNA) monitoring and immunotherapy for mismatch – repair deficient tumors – are expanding the therapeutic arsenal [12,22].

This retrospective observational cohort study examines TNT outcomes in patients with LARC treated at a national referral center using a sandwich TNT strategy. We hypothesized that TNT delivery and sequencing outcomes in routine clinical practice would be consistent with published trial benchmarks, and that carefully selected W&W with structured surveillance would offer a feasible organ – preservation option. We therefore conducted a retrospective cohort study to characterize TNT delivery, response endpoints (pCR, TRG, cCR with organ preservation), and survival outcomes in a consecutive series of LARC patients treated at our institution, and to contextualize these results against established TNT trial data and W&W registries.

## 2. Materials and Methods

### *2.1. Study Design and Patient Population*

This retrospective observational cohort study included consecutive adult patients who underwent the institutional sandwich TNT protocol [23] (induction multi-agent chemotherapy followed by long-course IMRT-SIB chemoradiotherapy with capecitabine and consolidation chemotherapy) for histologically confirmed rectal adenocarcinoma at the Institute of Oncology Ljubljana between January 2016 and December 2023 (Figure 1). The study followed STROBE (Strengthening the Reporting of Observational Studies in Epidemiology) recommendations for reporting observational research [24]. The study was conducted in accordance with the Declaration of Helsinki and approved by the institutional ethics committee; informed consent was waived due to retrospective analysis of de – identified data.

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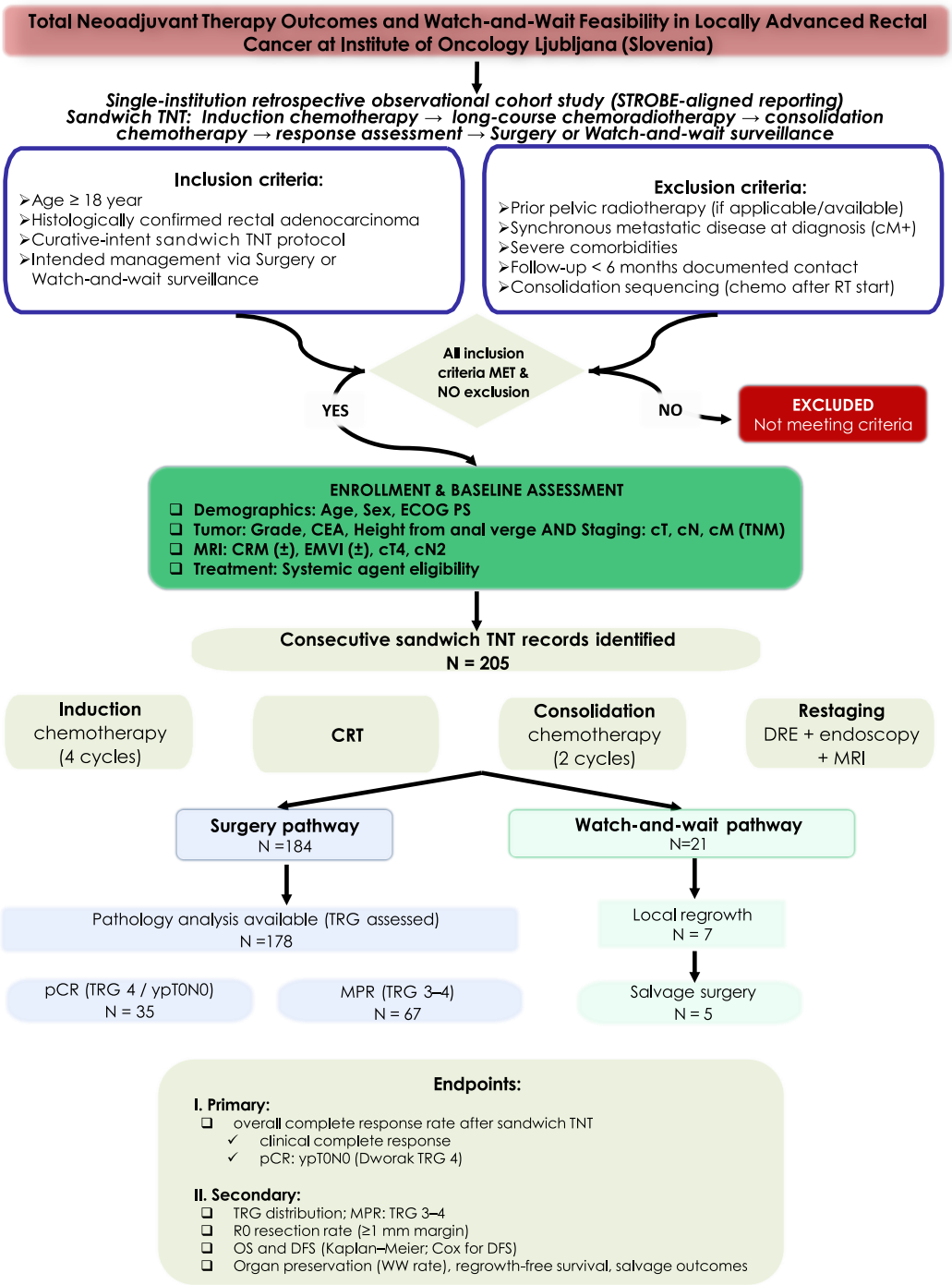


**Figure 1.** Flowchart of study protocol. CEA, carcinoembryonic antigen; cM/cM+, clinical M stage/clinical M1 (synchronous metastatic disease); cN, clinical nodal stage (N in TNM); CRM, circumferential resection margin; CRT, chemoradiotherapy; cT, clinical tumor stage (T in TNM); DFS, disease-free survival; DRE, digital rectal examination; ECOG PS, Eastern Cooperative Oncology Group performance status; EMVI, extramural venous invasion; MPR, major pathological response; MRI, magnetic resonance imaging; OS, overall survival; pCR, pathological complete response; R0, complete resection with negative margins (≥ 1 mm margin); TNM, tumor – node – metastasis classification; TNT, total neoadjuvant therapy; TRG, tumor regression grade; W&W , watch-and-wait; ypT0N0, pathological stage T0N0 after neoadjuvant therapy.

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*2.2. Eligibility Criteria*

Inclusion Criteria

- Histologically confirmed primary adenocarcinoma of rectum
- Clinical staging: cT3 – 4 and/or cN+ disease, cM0 (no distant metastases)
- Performance status: Eastern Cooperative Oncology Group (ECOG) 0 – 2
- Fitness for TNT: Adequate organ function to tolerate chemotherapy and radiotherapy
- Minimum of 6 months of follow – up available (unless death occurred earlier)

Exclusion Criteria

- Distant metastatic disease (cM1)
- Prior pelvic radiotherapy or systemic therapy for rectal cancer
- Pregnancy or nursing status
- Severe comorbidities precluding neoadjuvant therapy
- Non – evaluable/inadequate quality preoperative imaging
- Substantial missing key baseline staging data (define as missingness in ≥ 1 key MRI staging domain or > 20% missing among prespecified variables)

*2.3. Radiotherapy Delivery Treatment Protocol*

Preoperative radiotherapy was delivered using intensity-modulated radiation therapy with a simultaneous integrated boost (IMRT-SIB) according to the institutional protocol previously described by But-Hadzic et al. [23]. Patients underwent CT simulation in the supine position with a full-bladder protocol, with optional MRI fusion for target delineation. Target volumes were defined according to International Commission on Radiation Units and Measurements (ICRU) recommendations, with the gross tumor volume based on MRI and CT findings and clinical target volumes encompassing the mesorectum and regional lymphatic drainage areas [23,25]. Planning target volumes were generated using institutional margins accounting for organ motion and setup uncertainty.

Radiotherapy was delivered using IMRT with inverse planning and daily image guidance. The pelvic planning target volume received 41,8 Gy in 22 fractions, with a simultaneous integrated boost to the primary tumor to 46,2 Gy for cT3 tumors and 48,4 Gy for cT4 tumors. Treatment was administered in five fractions per week. Concomitant chemotherapy consisted of oral capecitabine (825 mg/m² twice daily) throughout the course of radiotherapy.

Detailed technical aspects of contouring, dose prescription, organ-at-risk constraints, and treatment delivery have been reported previously [23].

*2.4. Data Collection and Variables*

Data were extracted from electronic medical records into a standardized database including demographics, staging, treatment delivery, pathology, and outcomes. Baseline MRI variables included clinical T/N stage, CRM status, mrEMVI status, and tumor height from the anal verge. CRM involvement was defined as tumor ≤ 1 mm from the mesorectal fascia, consistent with commonly used MRI criteria [26]. mrEMVI was recorded on baseline MRI per radiologic criteria describing tumor signal within extramural vessels beyond the muscularis propria [27].

Radiotherapy related data was captured as total dose (Gy), fractionation, completion, and interruptions. Chemotherapy variables included regimen, intended and delivered cycles, dose reductions/delays, discontinuation and reason, and relative dose intensity. TNT sequencing was defined by the timing of chemotherapy initiation relative to radiotherapy.

Surgery variables included surgical procedure type and date. Pathology variables included ypT/ypN, resection margin status (R0/R1/R2), lymph node yield, LVI, PNI, tumor budding, microsatellite instability (when available), and tumor regression grade (TRG) using the Dworak 0 – 4 classification [28].

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For W&W surveillance, we recorded W&W entry, surveillance assessments, local regrowth events, and salvage surgery details.

### *2.5. Outcome Definitions*

#### Primary Endpoint

The primary endpoint was the overall complete response rate after sandwich TNT, defined as the proportion of patients achieving either a clinical complete response (cCR) at restaging or a pathological complete response (pCR) after surgery.

#### Key Secondary Endpoints

Key secondary endpoints were survival outcomes for the whole TNT cohort (surgery and W&W combined):

1. Overall survival (OS): time from diagnosis to death from any cause, with patients alive censored at last follow – up.
2. Disease – free survival (DFS): time from diagnosis to first documented disease recurrence (local, regional, or distant) or death from any cause, whichever occurred first, censored at last follow – up.

Additional prespecified outcomes were defined to characterise treatment response, organ preservation, and locoregional control:

1. Major pathological response (MPR): Dworak TRG 3 – 4 (good or complete regression) [28].
2. R0 resection rate: proportion of resections with all margins negative, defined as a minimum clearance ≥ 1 mm.
3. Organ preservation: W&W enrolment rate among the TNT cohort and durability of rectal preservation over time.
4. W&W regrowth: incidence, timing, and clinical/radiological characteristics of local regrowth in patients managed non-operatively after initial cCR. Local regrowth was defined as tumor reappearance at the primary site after cCR during non-operative management, distinct from post-resection local recurrence. Regrowth-free survival was calculated from entry into W&W protocol to first regrowth or censoring at last follow-up.
5. Salvage surgery outcomes: feasibility of salvage total mesorectal excision (TME) for regrowth, R0 resection rate after salvage, and DFS following salvage surgery
6. Post-resection local recurrence: crude incidence and landmark local pelvic recurrence rates among surgically treated patients. Because the exact date of local recurrence was not consistently documented in all cases, post-resection local control was evaluated using incidence proportions and landmark recurrence rates rather than formal local-recurrence-free survival curves

### *2.6. Statistical Analysis*

Continuous variables are presented as median (IQR), categorical variables as count (%). Baseline characteristics were compared using Wilcoxon rank – sum tests and Fisher exact tests.

pCR rates are presented with exact 95% CI using the Clopper – Pearson method. Overall survival and DFS are estimated using the Kaplan – Meier method with log – rank tests. Survival is presented at 2, 3, and 5 years with 95% CI.

Multivariable Cox proportional hazards models were used to identify DFS predictors; model discrimination assessed using Harrell's C – index [29]. Proportional hazards assumption tested using Schoenfeld residuals [30].

Watch – and – wait related outcome, regrowth – free survival is estimated using Kaplan – Meier methods. Regrowth incidence and time – to – regrowth are presented descriptively. Among patients with regrowth, salvage surgery rates, margins achieved, and subsequent DFS are reported.

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All p – values are two – sided, and p < 0,05 is considered statistically significant. No adjustment for multiple comparisons applied given the exploratory nature. R statistical software (version 4,3,2, R Core Team, R Foundation for Statistical Computing, Vienna, Austria) was utilized for the analysis.

*2.7. Ethics*

The Slovenian National Medical Ethical Committee (decision number: 0120-298/2019/5), Institutional Review Board (10/01/2019) and Institutional Ethical Committee (24/1/2019) approved the study. Due to the retrospective nature of the study, the need to obtain informed consent from participants was waived. All patient identifiers were removed, and analysis was conducted on de – identified data.

## 3. Results

*3.1. Study Cohort and Baseline Characteristics*

A total of 205 consecutive patients with rectal adenocarcinoma received induction – first approach TNT. Baseline characteristics are detailed in Table 1. Median age was 61 years (IQR: 52 – 67), 65% (n = 133) were male. Most patients presented with cT3 – 4 disease (98%, n = 200) and cN2 stage (60%, n = 124). CRM was threatened/involved in 61% (n = 122), and EMVI was positive in 66% (n = 120). Tumor grade was well/moderate in 87% and poor/undifferentiated in 13%. Among 205 patients, 184 (90%) underwent surgical resection, 21 (10%) were managed with W&W surveillance.

**Table 1.** Clinicopathological characteristics of the study cohort.

| Characteristic | Overall (N = 205)[1] | Surgery (N = 184)[1] | Watch – and – wait (N = 21)[1] |
|---|---|---|---|
| **Sex** | | | |
| Male | 133 (65%) | 121 (66%) | 12 (57%) |
| Female | 72 (35%) | 63 (34%) | 9 (43%) |
| **Age at diagnosis (years)** | | | |
| Median (Q1, Q3) | 61 (52, 67) | 61 (52, 67) | 63 (55, 68) |
| Min – Max | 32 – 80 | 32 – 80 | 33 – 77 |
| **ECOG performance status** | | | |
| 0 | 151 (74%) | 135 (73%) | 16 (76%) |
| 1 | 53 (26%) | 48 (26%) | 5 (24%) |
| ≥2 | 1 (0,5%) | 1 (0,5%) | 0 (0%) |
| **Baseline CEA (ng/mL)** | | | |
| Median (Q1, Q3) | 4 (2, 11) | 4 (2, 11) | 4 (2, 8) |
| Min – Max | 0 – 375 | 0 – 375 | 1 – 23 |
| **Tumor grade** | | | |
| G1 (Well) | 26 (15%) | 26 (17%) | 0 (0%) |
| G2 (Moderate) | 126 (73%) | 106 (69%) | 20 (100%) |
| G3 (Poor) | 13 (7,5%) | 13 (8,5%) | 0 (0%) |
| Mucinous / Undifferentiated | 8 (4,6%) | 8 (5,2%) | 0 (0%) |
| Unknown | 32 | 31 | 1 |
| **Distance from anal verge** | | | |
| 0 – 5 cm | 84 (41%) | 75 (41%) | 9 (43%) |
| 5,1 – 10 cm | 94 (46%) | 84 (46%) | 10 (48%) |
| 10,1 – 15 cm | 27 (13%) | 25 (14%) | 2 (9,5%) |
| **Clinical T stage (cT)** | | | |

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| | | | |
|---|---|---|---|
| 2 | 5 (2,4%) | 5 (2,7%) | 0 (0%) |
| 3 | 116 (57%) | 98 (53%) | 18 (86%) |
| 4 | 84 (41%) | 81 (44%) | 3 (14%) |
| **Clinical N stage (cN)** | | | |
| 0 | 14 (6,8%) | 10 (5,4%) | 4 (19%) |
| 1 | 67 (33%) | 58 (32%) | 9 (43%) |
| 2 | 124 (60%) | 116 (63%) | 8 (38%) |
| **MRI CRM threatened/involved** | **122 (61%)** | **107 (59%)** | **15 (71%)** |
| Missing | 4 | 4 | 0 |
| **MRI EMVI positive** | **120 (66%)** | **110 (68%)** | **10 (48%)** |
| Missing | 23 | 23 | 0 |

[1]n (%), [2]Fisher's exact test for categorical variables; Wilcoxon rank – sum test for continuous variables; CEA, carcinoembryonic antigen; CI, confidence interval; cCR, clinical complete response; cN, clinical nodal stage; cT, clinical T stage; CRM, circumferential resection margin (threatened/involved on baseline MRI); ECOG PS, Eastern Cooperative Oncology Group performance status; EMVI, extramural vascular invasion (positive on baseline MRI); IQR, interquartile range; LN, lymph node; MRI, magnetic resonance imaging.

Treatment compliance with the planned total neoadjuvant therapy regimen was high across the entire cohort. Radiotherapy was completed as planned in all patients, corresponding to a completion rate of 100% in the overall cohort as well as in both the surgically treated and W&W groups.

The majority of patients completed the planned systemic therapy, with 90% of patients in the overall cohort receiving all six planned chemotherapy cycles. Chemotherapy dose reductions were uncommon, and 93% of patients in the overall cohort completed treatment without dose reduction.

Chemotherapy discontinuation occurred in 8,8% of patients overall, including 8,7% in the surgery group and 9,5% in the watch-and-wait group. Discontinuation was most frequently related to treatment-related toxicity, accounting for 9,3% of cases in the overall cohort. No treatment discontinuations occurred due to patient preference. Overall, treatment adherence to the planned neoadjuvant regimen was high in both treatment pathways.

### *3.2. Pathologic Response and Surgical Outcomes*

Among 184 surgically resected patients, overall pCR rate (ypT0N0) was 19,3% (95% CI 13,9 – 25,9%).

Tumour regression grade (Dworak TRG 0 – 4) [28] distribution was as follows: TRG 0 (1,1%), TRG 1 (21,1%), TRG 2 (38,4%), TRG 3 (16,8%), and TRG 4 (19,5%). Major pathological response (MPR; TRG 3 – 4) was observed in 37,6% (N = 67) of resected patients. R0 resection (negative margins, ≥ 1 mm) was achieved in 94,5% (n = 173) of resected patients; R1 in 4,9% (n = 10), and R2 in one patient (0,5%). Lymphovascular invasion (LVI) was present in 23,2 (n = 42) and perineural invasion (PNI) in 15,9% (n = 29).

Univariable associations between baseline clinical/MRI features and major pathological response are presented in Table 2. These exploratory analyses evaluate whether MRI high – risk features (CRM involvement, EMVI, cT4, cN2) and performance status are associated with achieving MPR after sandwich TNT protocol and surgery.

**Table 2.** Univariable predictors of major pathologic response (MPR = TRG 3 – 4).

| | Descriptive | | | Logistic Regression | | | |
|---|---|---|---|---|---|---|---|
| **Characteristic** | **Overall N = 178[1]** | **TRG 1-2 N = 111[1]** | **TRG 3-4 N = 67[1]** | **N** | **OR** | **95% CI** | **p-value** |
| **Sex** | | | | **178** | | | |
| Male | 117 (66%) | 72 (65%) | 45 (67%) | | – | – | |

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| | | | | | | | |
|---|---|---|---|---|---|---|---|
| Female | 61 (34%) | 39 (35%) | 22 (33%) | | 0,90 | 0,47–1,71 | 0,8 |
| **ECOG PS** | | | | **178** | | | |
| 0 | 131 (74%) | 79 (71%) | 52 (78%) | | – | – | |
| 1 | 46 (26%) | 31 (28%) | 15 (22%) | | 0,74 | 0,35–1,48 | 0,4 |
| ≥ 2 | 1 (0,6%) | 1 (0,9%) | 0 (0%) | | 0,00 | | > 0,9 |
| **Histologic grade** | | | | **147** | | | |
| G1 (Well) | 24 (16%) | 12 (13%) | 12 (24%) | | – | – | |
| G2 (Moderate) | 103 (70%) | 69 (72%) | 34 (67%) | | 0,49 | 0,20–1,22 | 0,12 |
| G3 (Poor) | 12 (8,2%) | 8 (8,3%) | 4 (7,8%) | | 0,50 | 0,11–2,05 | 0,3 |
| Mucinous / Undifferentiated | 8 (5,4%) | 7 (7,3%) | 1 (2,0%) | | 0,14 | 0,01–0,98 | 0,089 |
| **CEA (pre)** | **4 (2-11)** | **5 (2-11)** | **4 (2-9)** | **178** | **1,00** | **0,99–1,01** | **> 0,9** |
| **Tumor height** | | | | **178** | | | |
| 0 – 5 cm | 74 (42%) | 47 (42%) | 27 (40%) | | – | – | |
| 5,1 – 10 cm | 80 (45%) | 47 (42%) | 33 (49%) | | 1,22 | 0,64–2,35 | 0,5 |
| 10,1 – 15 cm | 24 (13%) | 17 (15%) | 7 (10%) | | 0,72 | 0,25–1,89 | 0,5 |
| **cT stage** | | | | **178** | | | |
| 2 | 4 (2,2%) | 2 (1,8%) | 2 (3,0%) | | – | – | |
| 3 | 97 (54%) | 59 (53%) | 38 (57%) | | 0,64 | 0,07–5,55 | 0,7 |
| 4 | 77 (43%) | 50 (45%) | 27 (40%) | | 0,54 | 0,06–4,70 | 0,5 |
| **cN stage** | | | | **178** | | | |
| 0 | 9 (5,1%) | 6 (5,4%) | 3 (4,5%) | | – | – | |
| 1 | 55 (31%) | 41 (37%) | 14 (21%) | | 0,68 | 0,16–3,57 | 0,6 |
| 2 | 114 (64%) | 64 (58%) | 50 (75%) | | 1,56 | 0,39–7,69 | 0,5 |
| **CRM+** | | | | **178** | | | |
| No | 71 (39%) | 42 (39%) | 28 (42%) | | – | – | |
| Yes | 104 (58%) | 66 (59%) | 38 (57%) | | 0,89 | 0,48–1,66 | 0,7 |
| **EMVI+** | | | | **178** | | | |
| No | 50 (28%) | 31 (31%) | 19 (34%) | | – | – | |
| Yes | 105 (59%) | 68 (61%) | 37 (55%) | | 0,78 | 0,42–1,44 | 0,4 |
| **Dose reduction** | | | | **178** | | | |
| No reduction | 165 (93%) | 102 (92%) | 63 (94%) | | – | – | |
| Oxaliplatin reduced | 6 (3,4%) | 3 (2,7%) | 3 (4,5%) | | 1,62 | 0,29–8,98 | 0,6 |
| Capecitabine reduced | 3 (1,7%) | 3 (2,7%) | 0 (0%) | | 0,00 | | > 0,9 |
| Both reduced | 4 (2,2%) | 3 (2,7%) | 1 (1,5%) | | 0,54 | 0,03–4,32 | 0,6 |

[1]n (%); Median (Q1-Q3); Abbreviations: CI = Confidence Interval, OR = Odds Ratio, MPR, major pathologic response (TRG 3 – 4 Dworak tumor regression grade [28]).

*3.3. Organ Preservation and Watch – and – Wait Outcomes*

Of 205 patients treated with a sandwich TNT protocol, 21 (10,2%) were ultimately managed with the W&W strategy after restaging. W&W selection was restricted to patients with a clinical complete or near – complete response at the primary tumor site according to the predefined criteria. W&W outcomes are detailed in Table 3. Among 21 patients selected for W&W, the median age was 63 years; 57,1% were male.

Over a median follow – up of 4,96 years from W&W protocol entry, local regrowth occurred in 7 (33,3%) (95% CI 14,6 – 57,0%). Median time to regrowth was 10,8 months (range 6,1 – 42,5 months). Regrowth – free survival at 1/2/3/5 years was 68,8% (95% CI: 49,4 – 95,7%), 62,5% (42,8 – 91,4%), 62,5% (42,8 – 91,4%), and 55,6% (35,6 – 86,6%), respectively (Table 3 and Figure 2). Among patients with regrowth, 5 (71,4%) underwent salvage surgery. R0 was achieved in all patients.

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**Table 3.** Watch – and – wait (W&W) management outcomes. Clinical characteristics and oncologic results for 21 patients selected for W&W following total neoadjuvant therapy (TNT).

| Metric | Value |
|---|---|
| W&W cohort size | 21 |
| Median age (IQR), years | 63 (55 – 68) |
| Male sex, n (%) | 12/21 (57,1%) |
| Median follow – up from W&W entry (IQR), years | 4,96 (2,50 – 5,57) |
| Local regrowth, n/N (%) | 7/21 (33,3%) |
| Local regrowth, 95% CI | 14,6 – 57,0% |
| Median time to regrowth (range), months | 10,8 (6,1 – 42,5) |
| Regrowth – free survival at 1/2/3/5 years (95% CI) | 1 – year: 68,8% (49,4 – 95,7);<br>2 – year: 62,5% (42,8 – 91,4);<br>3 – year: 62,5% (42,8 – 91,4);<br>5 – year: 55,6% (35,6 – 86,6) |
| Salvage surgery among regrowth, n/N (%) | 5/7 (71,4%) |
| R0 among salvage resections, n/N (%) | 5/5 (100,0%) |
| Surgery – free survival at 5 years (95% CI) | 73,10% (55,0 – 97,20%) |

W&W, watch – and – wait; IQR, interquartile range; CI, confidence interval; R0, microscopically margin – negative resection.

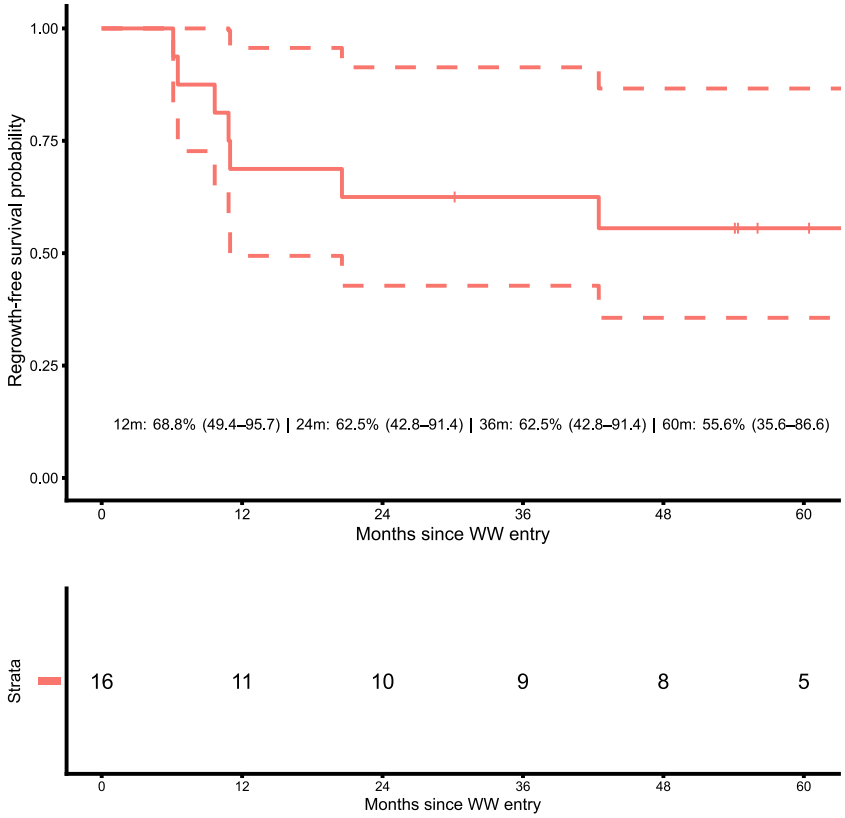


**Figure 2.** Regrowth – free survival among patients managed with watch – and – wait (W&W) strategy after sandwich total neoadjuvant therapy (TNT) protocol. Kaplan – Meier curve with 95% confidence intervals demonstrates Regrowth – free survival probabilities of 68,8% (95% CI: 49,4 – 95,7%) at 12 months, 62,5% (42,8 –

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91,4%) at 24 and 36 months, and 55,6% (35,6 – 86,6%) at 60 months. The risk table below curve shows the number at risk at each time point. Time zero is defined as entry into W&W protocol following clinical response assessment by MRI and endoscopy. W&W, watch – and – wait.

### *3.4. Survival Outcomes*

#### 3.4.1. Overall Survival

With a median follow – up of 5,04 years (95% CI: 4,86–5,43) from diagnosis, the observed follow-up time ranged from 0,71 to 9,01 years. There were 36 (17%) deaths among all 205 patients. Overall survival at 2, 3, and 5 years was 94,2% (95% CI: 91,0%–97,4%), 86,7% (95% CI: 82,0%–91,6%), and 81,1% (95% CI: 75,5%–87,2%), respectively (Table 4).

When stratified by management pathway OS at 2, 3, and 5 years in the resected cohort was 94,0% (95% CI 90,6–97,5%), 86,8% (95% CI 81,8–92,0%), and 80,3% (95% CI 74,2–87,0%). In the W&W cohort, corresponding OS estimates were 95,0% (95% CI 85,9–100,0%), 84,0% (95% CI 68,8.–100,0%), and 84,0% (95% CI 68,8–100,0%) (Table 4). OS did not differ significantly between the resected and W&W pathways (log-rank p = 0,69, Figure 3).

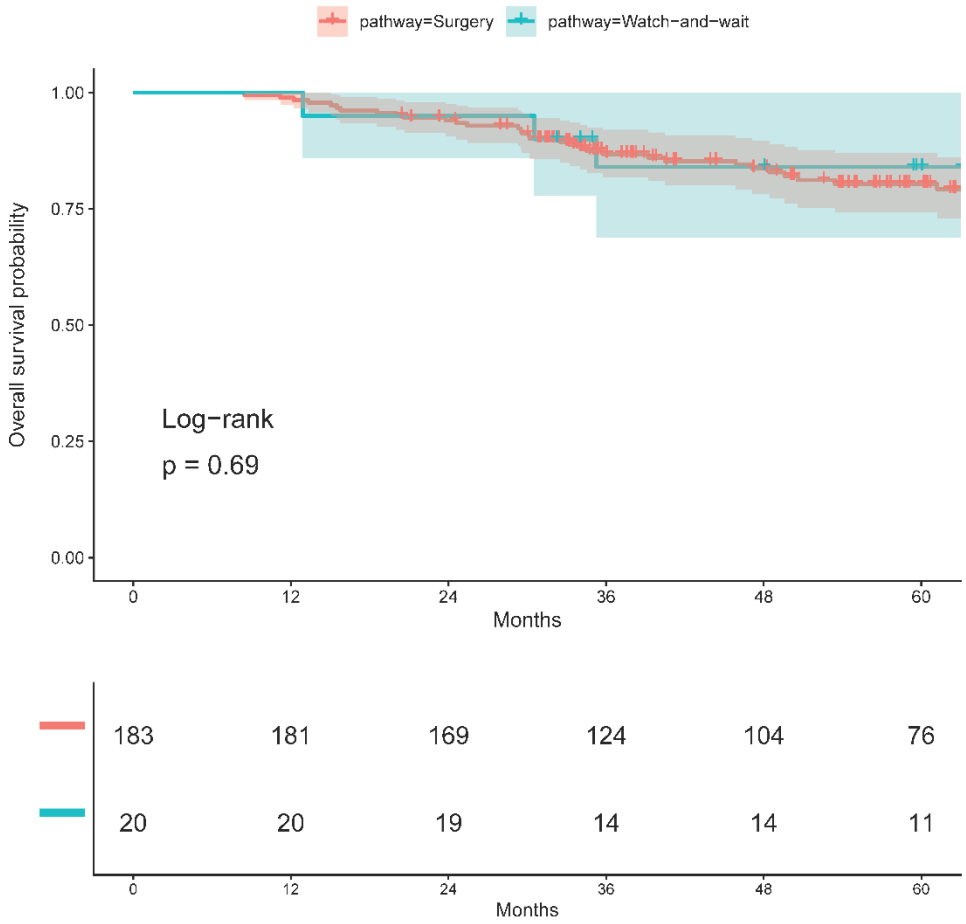


**Figure 3.** Overall survival (OS) by pathway. Kaplan – Meier curves comparing OS between watch – and – wait (W&W) and radical surgery (RS) cohorts following sandwich total neoadjuvant therapy (TNT) for locally advanced rectal cancer (LARC). Log – rank p – value shown.

**Table 4.** Overall survival estimation at prespecified time points by treatment pathway.

| | **Overall survival estimation at prespecified time points (%, 95%CI)** | | |
|---|---|---|---|
| | **Month 24** | **Month 36** | **Month 60** |
| **Whole cohort** | **94,2% (91,0–97,4),** | **86,7% (82,0–91,6)** | **81,1% (75,5–87,2)** |

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| | | | |
|---|---|---|---|
| **Surgery** | 94,0% (90,6 – 97,5) | 86,8% (81,8 – 92,0) | 80,3% (74,2 – 87,0) |
| **W&W** | 95,0% (85,9 – 100,0) | 84,0% (68,8 – 100,0) | 84,0% (68,8 – 100,0) |

W&W, watch – and – wait.

3.4.2. Disease – Free Survival

Among 205 patients, 56 experienced a DFS event. Median follow-up was 64,3 months (95% CI: 60,1–69,9). Estimated DFS was 93,7% (95% CI: 90,4%–97,1%) at 2 years, 84,1% (95% CI: 79,1%–89,4%) at 3 years, and 75,2% (95% CI: 69,0%–82,0%) at 5 years (Figure 4, Panel A).

Among 184 resected patients, disease – free survival at 2, 3, and 5 years was 93,5% (95% CI 90 – 97,1%), 83,5% (95% CI 78,1 – 89,2%), and 73,5% (95% CI 66,7 – 80,9%), respectively with median not reached. Figure 4 (Panel B) depicts the overall Kaplan – Meier DFS probability curve for these patients. Figure 4 illustrates DFS stratified by CRM status (Panel C), EMVI status (Panel D), and surgical margin status (Panel E). CRM – positive patients exhibited inferior 3 – year DFS compared to CRM – negative patients (77% vs 94%; 70 vs 54 at risk; log – rank p = 0,003). EMVI – positive status was associated with worse 3 – year DFS (79% vs 98%; 69 vs 39 at risk; p < 0,001). Surgical margin status showed the strongest prognostic impact, with R0 resections achieving 89% 3 – year DFS compared to 18% for R1 – R2 resections (124 vs 2 at risk; p < 0,001).

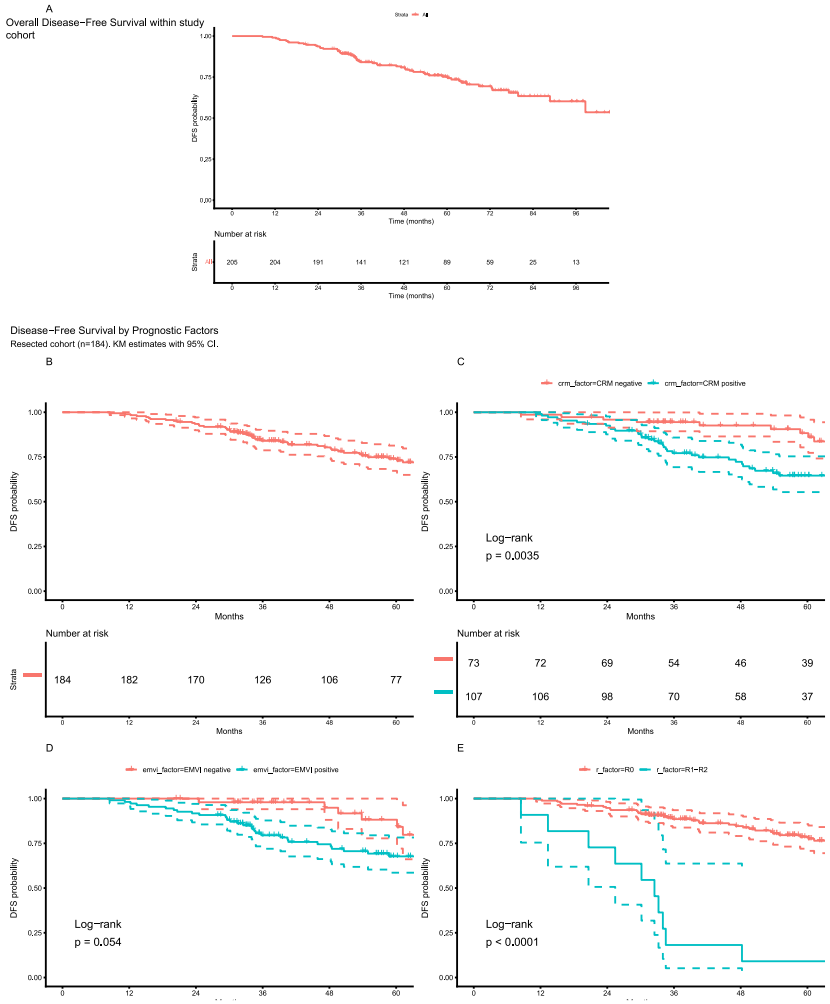


**Figure 4.** Overall disease – free survival (DFS) probability after sandwich TNT (Panel A), DFS within resected patients (N = 184, Panel B) and disease – free survival probability stratified by circumferential resection margin (CRM) status (Panel C), extramural vascular invasion (EMVI) status (Panel D) and by surgical margin (R0 vs R1 – R2 resection) status (Panel E).

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In multivariable Cox regression analysis (Figure 5), R0 resection was associated with a markedly lower risk of DFS events (R1/2 vs. R0 HR 6,06, 95% CI 1,99 – 18,42, p = 0,001), consistent with the strong adverse effect of any margin involvement. MRI CRM positivity independently worsened DFS (HR 3,11, 95% CI 1,53 – 6,33, p = 0,002), and MRI EMVI positivity showed a near – significant association with poorer DFS (HR 1,97, 95% CI 1,00 – 3,91, p = 0,051). Age showed a borderline effect, with a 33% relative increase in hazard per 10 – year increment (HR 1,33, 95% CI 0,99 – 1,79, p = 0,054). Clinical cT4 and cN2 stage did not significantly influence DFS after adjustment (cT4: HR 0,80, 95% CI 0,44 – 1,46, p = 0,467; cN2: HR 0,79, 95% CI 0,44 – 1,42, p = 0,436).

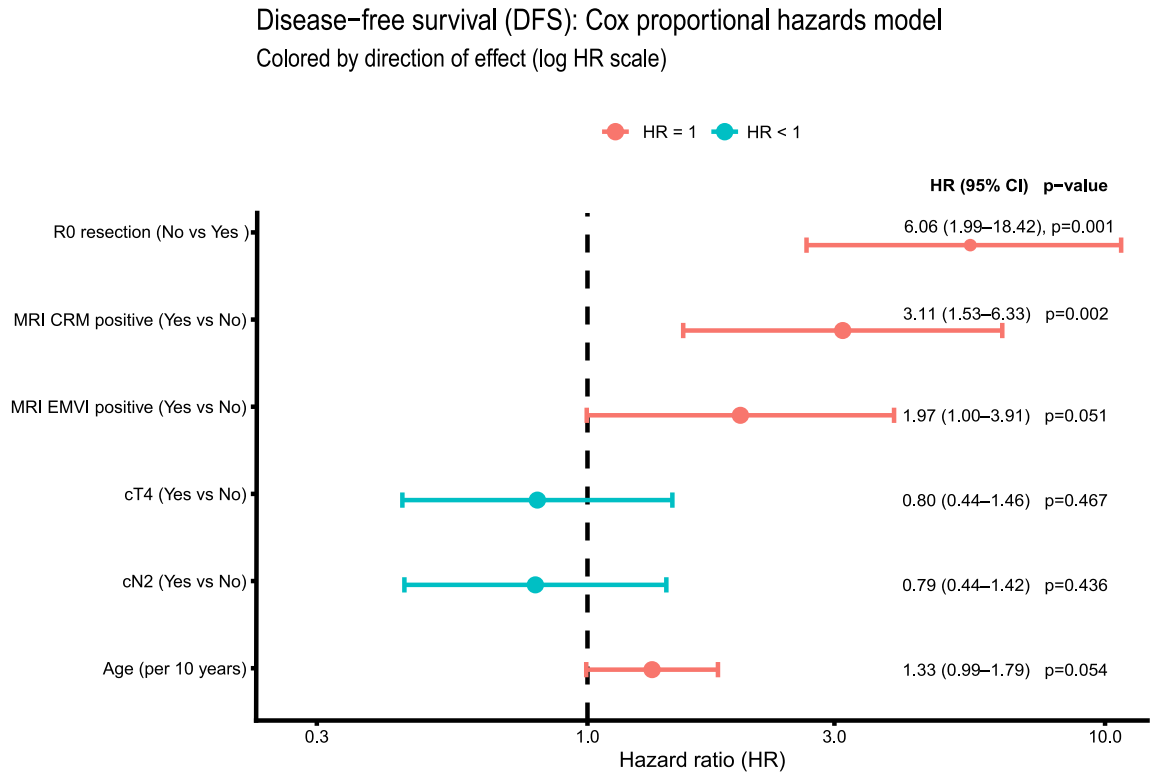


**Figure 5.** Multivariable Cox model for disease – free survival (DFS) in resected patients. Forest plot displaying adjusted hazard ratios (HRs) with 95% confidence intervals for MRI – based and pathological prognostic factors, plotted on a logarithmic scale. A vertical dashed line indicates the null value (HR = 1,0). Predictors to the right of the line (HR > 1) are associated with higher risk of DFS events, whereas those to the left (HR < 1) are associated with lower risk. Model concordance index was 0,723 (95% CI 0,652 – 0,794), with significant global likelihood ratio, Wald, and score tests (all p≤0,003). DFS, disease – free survival; HR, hazard ratio; CI, confidence interval; CRM, circumferential resection margin; EMVI, extramural vascular invasion; cT4, clinical T4 stage (tumor invades other organs/structures per AJCC staging); cN2, clinical N2 stage (≥ 4 regional lymph node metastases per AJCC staging); R0, microscopically margin – negative resection.

### 3.4.3. Local Recurrence

Local recurrence is reported as an incidence proportion with exact 95% confidence intervals, and as landmark incidence among patients with at least 2, 3, and 5 years of follow – up. Local recurrence occurred in 17 (9,3%) of resected patients (95% CI 5,5 – 14,5). Among patients with ≥ 24, ≥ 36, and ≥ 60 months follow – up, local recurrence rates were 8,3%, 4%, and 2,7%, respectively.

Incidence appeared higher in patients with CRM+, EMVI+, cT4, or cN2 disease, consistent with baseline risk features.

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3.4.4. Exploratory Comparison of Complete Responders – Surgical pCR Versus cCR Under Watch – and – Wait

Among patients achieving complete response, 35 had a pathological complete response (pCR, ypT0N0) after surgery and 21 were managed non – operatively with W&W) following a cCR at restaging restaging restaging.

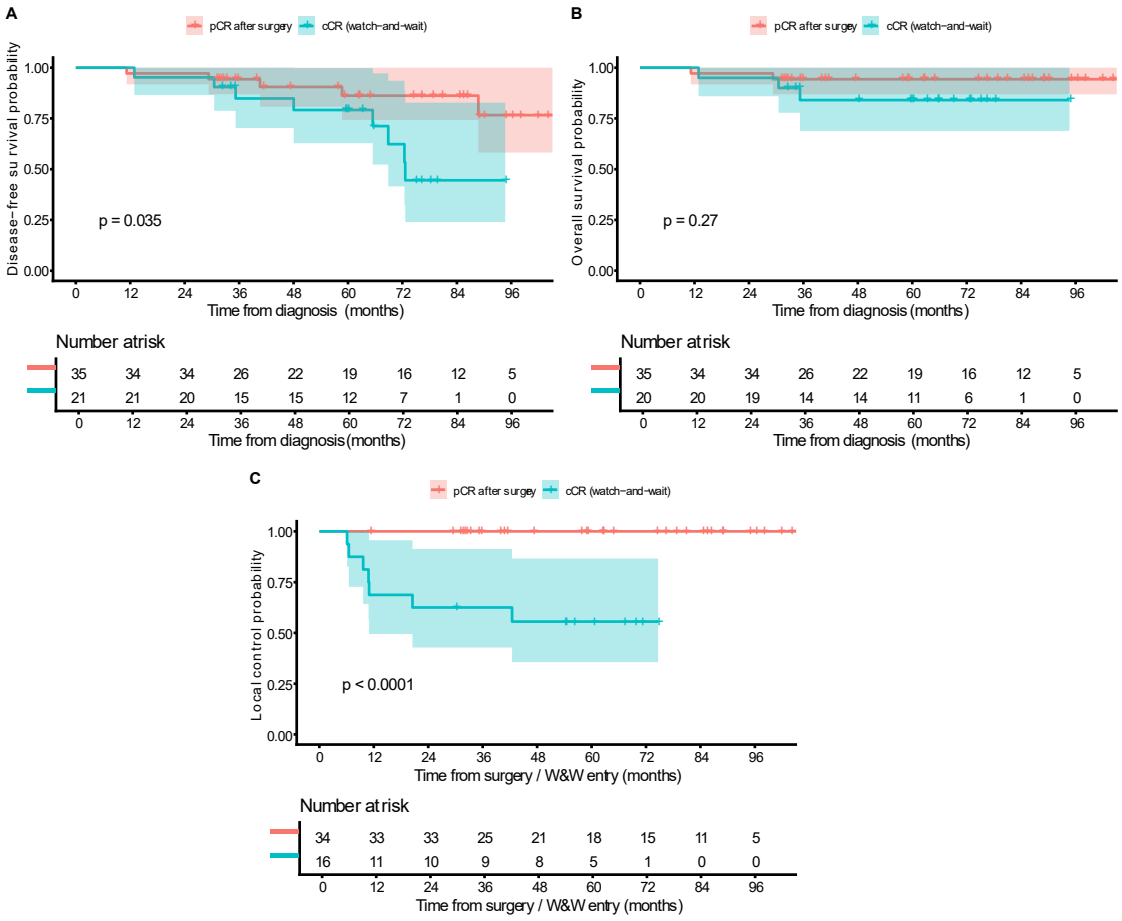


**Figure 6.** Exploratory comparison of complete responders: surgical pathological complete response (pCR) versus clinical complete response (cCR) managed with watch-and-wait (W&W). Panel A: disease-free survival (DFS) from diagnosis among complete responders, comparing patients with pCR treated with radical surgery versus those with cCR managed with watch-and-wait. Panel B: overall survival (OS) from diagnosis in the same subgroups. Panel C: cumulative incidence of local failure, showing absence of post-resection local recurrence in pCR/surgery patients and the pattern of local regrowth in cCR/ W&W patients. Abbreviations: pCR, pathological complete response; cCR, clinical complete response; W&W, watch-and-wait.

DFS from diagnosis was significantly higher in the pCR/surgery group compared with the cCR/W&W group (Figure 6A; log – rank p = 0,035). 3 – year DFS was 94,3% (95% CI 86,9 – 100) in the pCR group and 84,8% (95% CI 70,3 – 100) in the cCR/W&W group. In contrast, OS did not differ significantly between the two strategies, with high long – term survival observed in both cohorts (Figure 6B; log – rank p = 0,27). At 5 years, OS was was 94,3% (95% CI 86,9 – 100) in the pCR group and 84% (95% CI 68,8 – 100) in the cCR/W&W group. Differences were most pronounced for local control. No post-resection pelvic recurrences were observed among patients with pCR after surgery, whereas patients managed with W&W experienced local regrowth events (Figure 6C, log – rank p < 0,0001).

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## 4. Discussion

Sandwich total neoadjuvant therapy (TNT) has increasingly been adopted to improve systemic control, maximize delivery of planned chemotherapy, and increase tumor response in LARC, thereby expanding the pool of patients eligible for organ-preserving strategies. This retrospective study of consecutive cohort of 205 patients with LARC treated uniformly with a sandwich TNT strategy provides a pragmatic "implementation – level" assessment of outcomes achievable in routine practice within a European referral center. Across the entire cohort, the sandwich TNT approach yielded an overall complete response rate of 29,5% (pCR 19,3%, cCR 10,2%), major pathological response in 37,6%, and R0 resection in 94,5% of operated patients. Five-year OS and DFS for the whole TNT cohort were 81,1% (95% CI: 75,5%–87,2%) and 75,2% (95% CI: 69,0%–82,0%), respectively, and post-resection local recurrence remained below 10%. These whole-cohort endpoints are clinically informative because they reflect the composite effectiveness of a protocol that includes both radical surgery and a structured watch-and-wait (W&W ) pathway, mirroring contemporary practice where organ preservation is increasingly integrated into TNT-based management [31].

Our study results demonstrate substantial pathological stage migration, acceptable long – term oncologic outcomes, and the feasibility of selective W&W management. By integrating clinical – to – pathological stage shift, pathological response, and survival analyses, this study provides mechanistic insight into how TNT modifies disease burden while also delineates the limits of response in anatomically and biologically high – risk disease.

### *4.1. Tumour Response, Stage Migration and Survival in a High – Risk Cohort*

When interpreted in isolation, a pCR rate around 19% may appear modest relative to some contemporary TNT trials such as RAPIDO and PRODIGE-23, in which intensified neoadjuvant strategies produced higher pCR rates in broadly selected stage II–III populations [7,9,12]. However, the baseline risk profile in the present cohort is considerably more adverse than in most published randomised series: 61% had MRI-threatened or involved circumferential resection margin (CRM), 66% were EMVI-positive, 60% had cN2 nodal disease, and 41% had cT4 disease. MRI-defined CRM involvement and EMVI are well-established adverse prognostic features and can outperform TNM staging in predicting oncologic outcomes, supporting the clinical relevance of reporting baseline MRI risk composition when benchmarking TNT results. In this context, the observed proportion of patients downstaged to ypT0 – 2 and/or ypN0, major pathological response rate (Dworak TRG 3–4; 37,6%) and high R0 resection rate (94,5%) indicate clinically meaningful tumor regression in an exceptionally high-risk population.

Importantly, the whole-cohort 5-year DFS of 75,2% and OS of 81,1% are comparable to long-term TNT trial benchmarks, suggesting that the sandwich sequencing used here can deliver durable disease control in high-risk routine practice. This is aligned with the broader TNT evidence base demonstrating improvements in systemic control and DFS compared with conventional approaches, even though trial populations and treatment platforms differ.

Large phase III TNT trials such as RAPIDO and PRODIGE – 23 have demonstrated improvements in distant metastasis and DFS compared with conventional chemoradiotherapy followed by selective adjuvant chemotherapy, but in cohorts with somewhat different case – mix and staging criteria [7,9,12]. In PRODIGE – 23, induction mFOLFIRINOX followed by chemoradiotherapy and surgery yielded a pCR rate of 27,5%, but enrolled patients with cT3 – 4 M0 disease and lower MRI – defined risk than in the present series [11,12]. RAPIDO, which used short – course radiotherapy followed by consolidation chemotherapy before total mesorectal excision (TME), reported a substantial increase in pCR and a reduction in distant metastases at the cost of a slightly higher local relapse risk in long – term follow – up [9].

Within this broader TNT landscape, our results suggest that a sandwich protocol can yield pCR and DFS outcomes comparable to those reported in randomized trials, despite differences in

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radiotherapy regimen, chemotherapy backbone and substantially higher MRI – defined risk at baseline.

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*4.2. Persistent Prognostic Impact of MRI – Defined Anatomy and Margins*

Despite the overall efficacy of TNT in inducing tumor regression, our data highlight that pretreatment MRI risk factors and surgical margin status remain critical prognostic determinants. Patients whose tumors threatened or breached the mesorectal fascia on MRI had significantly worse outcomes, even after TNT and surgery. In our multivariable Cox analysis, an MRI-positive CRM was associated with more than a tripling of the hazard of relapse (HR 3,11, 95% CI 1,53–6,33), consistent with the MERCURY study findings that MRI-predicted margin involvement portends worse DFS and higher local recurrence [14]. Likewise, EMVI positivity was associated with a near-doubling of relapse risk (HR ~1,97), consistent with prior reports linking EMVI to distant metastases and poor survival [32,33].

Perhaps most striking is the influence of surgical margin quality. Patients undergoing an R1/R2 resection in our cohort had markedly inferior DFS compared to those with R0 resection (3 – year DFS 18% vs 89%), corresponding to a 6 – fold increased hazard of recurrence for incomplete resections. All 36 recorded deaths in our study occurred in patients with R1/R2 or other high – risk features, underscoring that clear margins (R0) are imperative for cure. These results emphasize that even in the TNT era, precise MRI staging and high – quality surgery are essential. We advocate upfront MRI risk stratification (e.g. for CRM, EMVI, tumor deposits) to guide therapy intensity and surgical planning. Patients with threatened CRM or bulky disease should be referred to experienced surgeons, as achieving an R0 resection can dramatically improve outcomes (DFS ~ 73% at 5 years in our R0 patients). In summary, TNT downstages disease but does not abrogate the prognostic importance of baseline risk factors or surgical technique. Ongoing refinements in MRI (e.g. tumor regression grading on restaging MRI) and incorporation of biomarkers may further inform which patients need augmented therapy beyond standard TNT.

*4.3. Sandwich TNT Protocol in Contemporary Sequencing Debates*

The optimal sequencing of total neoadjuvant therapy for LARC remains an area of active investigation. Contemporary randomized trials have primarily compared induction chemotherapy followed by chemoradiotherapy (CRT) with CRT followed by consolidation chemotherapy, rather than formally testing more complex schedules. Across these studies, TNT, irrespective of sequence, has consistently improved pathological response and reduced distant failure compared with conventional long – course CRT followed by selective adjuvant chemotherapy, as demonstrated in RAPIDO [9], PRODIGE – 23 [12] and related trials, and TNT is now endorsed by The European Society for Medical Oncology and other international guidelines for MRI – defined high – risk stage II – III rectal cancer [34]. More recently, the CAO/ARO/AIO – 12 and OPRA trials suggested that up – front CRT followed by consolidation chemotherapy may yield higher pCR and sustained cCR/organ preservation rates than induction – first TNT, without compromising disease – free survival, and has therefore become the preferred sequence in many centres when organ preservation is a priority [10,17]. Indeed, OPRA achieved a 3 – year organ preservation rate of 52% with consolidation – chemotherapy TNT, versus 41% with induction – first (p = 0,01), while 5 – year DFS was approximately 71% in both arms (p = 0,68). Based on these data, many centers now favor upfront chemoradiation then chemotherapy when the goal is non – operative management [35].

A distinctive component of our cohort is the radiotherapy delivery regimen, consisting of long-course hypofractionated IMRT-SIB delivering 41,8 Gy to the elective pelvic volume with a boost to 46,2 Gy for cT3 and 48,4 Gy for cT4 tumors in 22 fractions with concurrent capecitabine. This schedule differs from conventional long-course CRT fractionation commonly used across major TNT trials, and there are limited datasets integrating this exact IMRT-SIB fractionation into a sandwich TNT framework. By extending IMRT-SIB–based chemoradiotherapy into a large real-world sandwich TNT cohort with long follow-up, the present study helps address a practical evidence gap, namely whether this non-standard fractionation can achieve outcomes comparable to established neoadjuvant CRT schedules when embedded within modern TNT. This hybrid approach, delivered

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uniformly to all patients, attempts to capture the benefits of both sequences: early systemic therapy to tackle micrometastases, plus additional chemotherapy after radiation to maximize tumor regression prior to surgery. Phase II studies of similar sandwich regimens have reported pCR rates around 30 – 40% [36], exceeding those of induction regimens, although randomized evidence is sparse. Notably, delivering chemotherapy both before and after CRT did not appear to compromise tolerability in our series – 90% of patients completed all planned cycles.

While direct comparisons between sequencing strategies await prospective trials, our real – world data suggest that a sandwich approach can achieve oncologic outcomes on par with induction – only or consolidation – only TNT. This may be particularly relevant for centers or patients aiming to balance maximal tumor response with early systemic therapy. Moving forward, randomized studies (or pooled analyses) formally comparing mixed – sequence TNT to single – sequence regimens would help determine if outcomes can be further optimized by such adaptive scheduling.

### *4.4. Watch – and – Wait Outcomes and Salvageability*

Within the whole TNT cohort, 10,2% of patients were managed by W&W after achieving cCR, which represents a pragmatic organ-preservation rate in a cohort dominated by MRI high-risk features.

Two – thirds of our W&W patients were able to avoid surgery long – term, and salvage TME was successful in 100% of those who did recur. This is consistent with the OPRA trial and IWWD registry, which reported high salvage rates and no survival penalty for W&W [18,37]. Thus, for patients desiring organ preservation and meeting strict response criteria, W&W is a reasonable option. It must be coupled with rigorous surveillance. Patients should also be counseled that in real – world settings, up to 40 – 50% may ultimately require surgery due to tumor regrowth or indeterminate findings. Notably, OPRA demonstrated that the majority of regrowth events occur within the first 2 – 3 years, underscoring the importance of close early follow – up rather than regrowth rate alone as the primary safety metric [10,17].

Taken together, our W&W results support feasibility after sandwich TNT, while the regrowth rate underscores the need for strict selection criteria and intensive surveillance, particularly in MRI high-risk patients.

### *4.5. Limitations and Strengths*

This study has several limitations. It is a single – institution retrospective analysis, which carries inherent risks of selection bias and unmeasured confounding. Further on, the choice of W&W was not randomized and was influenced by clinical judgment, which could bias comparisons. The sample size of the W&W cohort (n = 21) was relatively small, limiting statistical power to detect minor differences and to perform extensive subgroup analyses (for instance, outcomes by cCR stringency or salvage timing). Additionally, pathologic tumor regression grading was performed by institutional pathologists without central review; while all used a standard Dworak TRG scheme, some inter – observer variability is possible. We also note that our median follow – up (~5 years for surviving patients) may be insufficient to capture very late recurrences, though > 90% of relapses in rectal cancer occur within 5 years. Despite these limitations, the study's strengths include a consecutive real – world cohort reflecting routine practice (enhancing generalizability), comprehensive MRI staging data for all patients, and mature survival follow – up exceeding 5 years. Moreover, the uniformity of the TNT protocol and surgical technique at our center provides a consistent treatment backdrop against which outcomes were assessed.

### *4.6. Clinical Implications and Future Directions*

The findings from this high – risk real – world cohort underscore the viability and effectiveness of sandwich TNT, integrating both induction and consolidation chemotherapy, in achieving favorable long – term oncologic outcomes. Outcomes in routine practice using a sandwich TNT

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protocol align with benchmarks from randomized TNT trials, indicating that induction – first sequencing with additional consolidation chemotherapy can achieve CR and survival results comparable to published series [11,12]. MRI – based risk stratification (CRM, EMVI) should guide treatment intensification and patient selection, as baseline tumor location and morphologic features remain independent prognostic factors even after TNT [14,16]. W&W with rigorous surveillance is a safe, effective organ – preservation strategy when structured MRI/endoscopy surveillance is available, multidisciplinary assessment confirms adequate cCR/near – cCR, patient commitment to surveillance is assured, and timely access to salvage surgery is guaranteed. However, patients must be counseled that regrowth occurs in 40 – 50% of W&W candidates in pragmatic settings, and careful baseline risk factor assessment is essential[10,17,18]. Resection margin status (R0 vs R1 – R2) is the dominant modifiable prognostic factor and should motivate specialized surgical referral and TME quality focus, particularly for high – risk disease (CRM+, EMVI+, cT4)[38–40]. Chemotherapy dose intensity (RDI > 85%) should be prioritized through proactive toxicity management and supportive care, as supported by literature on improved survival[41,42]. Emerging therapeutic strategies – including selective radiotherapy omission in favorable – risk patients, liquid biopsy monitoring, and immunotherapy for mismatch – repair deficient tumors – warrant integration into future TNT protocols to further personalize treatment and improve outcomes[19–22,43].

## 5. Conclusions

The results of this single-institution real-world cohort support several practice – relevant conclusions. Sandwich TNT delivered with an IMRT-SIB chemoradiotherapy backbone was feasible in routine practice for MRI-defined high-risk LARC and achieved durable long-term outcomes (5-year OS and DFS) together with high quality surgical outcomes. A carefully selected W&W strategy enabled meaningful organ preservation, with 5-year surgery-free survival of 73,1%, but regrowth occurred in one-third of patients, emphasizing that W&W should be reserved for strictly defined cCR/near-cCR responders and must be coupled with structured MRI/endoscopic surveillance and guaranteed access to timely salvage surgery. Baseline MRI risk features (CRM involvement, EMVI) and surgical margin status remained the dominant predictors of DFS, highlighting that high – quality TME and management in experienced tertiary centres continue to be critical determinants of outcome, even in the TNT era. Collectively, these pragmatic data support the feasibility of sandwich TNT implementation in routine practice and point to key priorities for future research, including prospective sequencing comparisons, integration of molecular and imaging biomarkers, and more refined, risk-adapted organ-preservation strategies for patients with LARC.

**Author Contributions:** Conceptualization, M.R. and V.V.; methodology, M.R. and V.V.; validation, V.V., F.A., A.J.P., P.K., I.O. and A.Š.E.; investigation, M.R. and V.V.; resources, M.R. and V.V.; writing—original draft preparation M.R. and V.V.; supervision, V.V., F.A., A.J.P., P.K., I.O. and A.Š.E. All authors have read and agreed to the published version of the manuscript.

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**Institutional Review Board Statement:** The study was conducted in accordance with the Declaration of Helsinki, and approved by the Slovenian National Medical Ethical Committee (decision number: 0120-298/2019/5), Institutional Review Board (10/01/2019) and Institutional Ethical Committee (24/1/2019).

**Informed Consent Statement:** Due to the retrospective nature of the study the need to obtain informed consent from participants was waived in accordance to the vote of the Slovenian National Medical Ethical Committee (decision number: 0120-298/2019/5), Institutional Review Board (10/01/2019) and Institutional Ethical Committee (24/1/2019).

**Data Availability Statement:** Data is contained within the manuscript. Further information about the data may be provided on request.

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**Conflicts of Interest:** The authors declare no conflicts of interest.

## Abbreviations

The following abbreviations are used in this manuscript:

| | |
|---|---|
| AJCC | American Joint Committee on Cancer |
| CAO-ARO-AIO | German Rectal Cancer Study Group acronym used in trial name (CAO/ARO/AIO-12) |
| CAPOX | Capecitabine plus oxaliplatin |
| CEA | Carcinoembryonic antigen |
| CI | Confidence interval |
| cCR | Clinical complete response |
| cM / cM+ | Clinical M stage / clinical M1 (synchronous metastatic disease) |
| cN | Clinical nodal stage (TNM) |
| cT | Clinical tumor stage (TNM) |
| CR | Complete response |
| CRM | Circumferential resection margin |
| CRT | Chemoradiotherapy |
| ctDNA | Circulating tumor DNA |
| DFS | Disease-free survival |
| DRE | Digital rectal examination |
| ECOG PS | Eastern Cooperative Oncology Group performance status |
| EMVI | Extramural vascular/venous invasion |
| EORTC | European Organisation for Research and Treatment of Cancer |
| ERID-KSOPKR | Institutional Review Board identifier (as written) |
| ERIDEK | Institutional Ethical Committee identifier (as written) |
| ESMO | European Society for Medical Oncology |
| FOLFIRINOX | Fluorouracil, leucovorin, irinotecan, oxaliplatin combination regimen |
| FOLFOX6 / mFOLFOX6 | (Modified) FOLFOX6 regimen (as cited) |
| HR | Hazard ratio |
| ICRU | International Commission on Radiation Units and Measurements |
| IMRT | Intensity-modulated radiation therapy |
| IMRT-SIB | Intensity-modulated radiation therapy with simultaneous integrated boost |
| IQR | Interquartile range |
| IWWD | International Watch & Wait Database |
| LARC | Locally advanced rectal cancer |
| LN | Lymph node |
| LR | Local recurrence |
| LVI | Lymphovascular invasion |
| mFOLFIRINOX | Modified FOLFIRINOX |
| MPR | Major pathological response |
| MRI | Magnetic resonance imaging |
| mrEMVI | MRI-detected extramural vascular invasion |
| MSI | Microsatellite instability |
| OPRA | Organ Preservation in Rectal Adenocarcinoma trial acronym |
| OR | Odds ratio |
| OS | Overall survival |
| pCR | Pathological complete response |
| PNI | Perineural invasion |
| PRODIGE | French cooperative group acronym in trial name (PRODIGE-23) |
| PROSPECT | PROSPECT trial acronym |
| R0 / R1 / R2 | Resection margin status: negative / microscopic positive / macroscopic positive |
| RAPIDO | RAPIDO trial acronym |
| RDI | Relative dose intensity |
| RS | Radical surgery |
| SCRT | Short-course radiotherapy |

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| | |
|---|---|
| SIB | Simultaneous integrated boost |
| STROBE | Strengthening the Reporting of Observational Studies in Epidemiology |
| TD | Tumor deposits |
| TME | Total mesorectal excision |
| TNM | Tumor, nodes, and metastases classification system |
| TNT | Total neoadjuvant therapy |
| TRG | Tumour regression grade |
| W&W | Watch-and-wait |
| ypT0N0 | Pathological stage T0N0 after neoadjuvant therapy |
| 95% CI | 95% Confidence Interval |

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