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Original Research

Novel phase I trial design to evaluate the addition of cediranib or selumetinib to preoperative chemoradiotherapy for locally advanced rectal cancer: the DREAMtherapy trial

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Abstract ***Background:*** The DREAMtherapy (Dual REctal Angiogenesis MEK inhibition radiotherapy) trial is a novel intertwined design whereby two tyrosine kinase inhibitors (cediranib and selumetinib) were independently evaluated with rectal chemoradiotherapy (CRT) in an efficient manner to limit the extended follow-up period often required for radiotherapy studies.

Patients and methods: Cediranib or selumetinib was commenced 10 days before and then continued with RT (45 Gy/25#/5 wks) and capecitabine (825 mg/m^2 twice a day (BID)). When three patients in the cediranib 15-mg once daily (OD) cohort were in the surveillance period, recruitment to the selumetinib cohort commenced. This alternating schedule was followed throughout. Three cediranib (15, 20 and 30 mg OD) and two selumetinib cohorts (50 and 75 mg BID) were planned. Circulating and imaging biomarkers of inflammation/angiogenesis

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were evaluated.
Results: In case of cediranib, dose-limiting diarrhoea, fatigue and skin reactions were seen in the 30-mg OD cohort, and therefore, 20 mg OD was defined as the maximum tolerated dose. Forty-one percent patients achieved a clinical or pathological complete response (7/17), and 53% (9/17) had an excellent clinical or pathological response (ECPR). Significantly lower level of pre-treatment plasma tumour necrosis factor alpha (TNFα) was found in patients who had an ECPR. In case of selumetinib, the 50-mg BID cohort was poorly tolerated (fatigue and diarrhoea); a reduced dose cohort of 75-mg OD was opened which was also poorly tolerated, and further recruitment was abandoned. Of the 12 patients treated, two attained an ECPR (17%).
Conclusions: This novel intertwined trial design is an effective way to independently investigate multiple agents with radiotherapy. The combination of cediranib with CRT was well tolerated with encouraging efficacy. TNFα emerged as a potential predictive biomarker of response and warrants further evaluation.

1. Introduction

Complete surgical resection after chemoradiotherapy (CRT) is the optimal management for patients with locally advanced rectal cancers (LARCs), where the circumferential resection margin (CRM) is threatened by tumour, involved node or extramural vascular invasion (EMVI) within a millimetre of the mesorectal fascia. Disease control has been improved with chemotherapy, yielding higher rates of pathological complete response (pCR) than radiotherapy alone [1,2]. However, even with this strategy, only about 15% of patients achieve a pCR [1,2] with a reduced local recurrence rate and superior outcome [3]. Multiple studies have evaluated the efficacy of adding other cytotoxic agents [4], although many have been unsuccessful while significantly increasing toxicity [4–7].

Preclinical and some clinical data have shown that vascular endothelial growth factor (VEGF) inhibitors can normalise tumour vasculature and increase the efficacy of coadministered radiation [8–12]. However, previous studies with the monoclonal anti-VEGF antibody, bevacizumab, were associated with significant toxicity, leading to early termination of some trials [13–15]. In addition, there are theoretical concerns about the long half-life of bevacizumab and its potential subsequent impact on surgical wound healing and fistula formation. These concerns led us to evaluate a VEGF receptor tyrosine kinase inhibitor (TKI), cediranib [16], instead of a monoclonal antibody, in combination with CRT.

One of the difficulties encountered when carrying out early-phase drug trials in combination with radiotherapy is that the trial protocol mandates a protracted follow-up period to assess both the acute and late effects of radiation. To mitigate this issue, we designed a two-drug phase I trial in which each of the agents was evaluated in alternating cohorts in combination with CRT. Thus, in this trial, the period of toxicity monitoring for the cediranib CRT cohort was used to recruit patients to a CRT combination with a second agent, selumetinib, and vice versa. This efficient trial design allowed two drugs to be investigated in combination with CRT, removing the period of trial interruption that is traditionally incorporated into phase I evaluations of novel radiation combination regimens.

The second drug evaluated in this study, selumetinib (AZD6244, ARRY-142886), is a potent, selective, inhibitor of mitogen-activated protein kinase kinase (MEK), which regulates cell proliferation and survival [17]. Preclinical data suggest that selumetinib enhances the effect of radiotherapy when used in combination [18].

The main objective of this trial was to establish the maximum tolerated dose (MTD) of both cediranib and selumetinib in combination with standard CRT for rectal cancer. A comprehensive translational research protocol was incorporated aiming to identify potential predictive biomarkers of response. Here, we describe the results generated with this novel trial design and report the striking clinical activity seen when cediranib was combined with CRT.

2. Materials and methods

2.1. Patients

Patients with histologically confirmed rectal cancer were eligible. Patients were recruited from a single institution between September 2010 and December 2012. All patients had rectal tumours that threatened the CRM (tumour, involved node or EMVI $\leq$ 1 mm from the mesorectal fascia), or the patient had T3 tumours less than 5 cm from the anal margin in the absence of distant metastases. Tumour was assessed by magnetic resonance imaging (MRI) and triphasic computed tomography (CT) scans. The patients were aged 18 years or more and had an Eastern Cooperative Oncology Group

performance status of 0 or 1 and adequate organ function (absolute neutrophil count $\geq 1.5 \times 10^9$/l; platelet count $>100 \times 10^9$/l; serum bilirubin <1.5 x upper limit of normal (ULN); serum alkaline phosphatase [ALP] < 5 x ULN; serum transaminase [aspartate aminotransferase (AST) or alanine aminotransferase (ALT)] < 2.5 x ULN; creatinine clearance >50 mL/min calculated by the Cockcroft-Gault formula). The patients recruited to the selumetinib cohorts also had to have a somatic mutation of the KRAS gene as it was felt that this would maximise the potential efficacy of selumetinib. NRAS testing was not routinely tested at the time of trial recruitment. A subsequent protocol amendment was passed, and patients with KRAS wild-type tumours were also enrolled in the 2nd cohort.

2.2. Study design and treatment

2.2.1. Intertwined trial design

The trial investigated cediranib or selumetinib with CRT using an alternating cohort design, which used the mandated follow-up interval between cohorts to evaluate a new dose level of the other drug–CRT (NCT01160926) combination. The study gained the relevant ethical (MREC 09/H1008/136) and regulatory approvals (EUDRACT: 2009 - 016524–31). All patients granted informed consent to participate. The trial was sponsored by The Christie NHS Foundation Trust.

The first cohort incorporated cediranib 15 mg OD. When patients in this cohort completed treatment, the initial cohort of selumetinib 50 mg BID started (Fig. 1). This alternating cohort design continued until a dose limiting toxicity (DLT) was observed.

2.2.2. Experimental drugs: cediranib and selumetinib

The single agent cediranib or selumetinib was commenced 10 days before (day 10) the first dose of CRT and continued throughout CRT. Three doses of cediranib were planned: 15 mg, 20 mg and 30 mg OD. Three patients were recruited to each cohort. If a DLT was encountered, the cohort was expanded to include six patients. Two doses were planned for selumetinib: 50 mg and 75 mg BID, and a further reduced dose level (75 mg OD) was mandated if toxicity was observed with these cohorts.

2.2.3. Chemoradiotherapy

Pelvic radiotherapy was administered to a CT-planned volume at a dose of 45 Gy in 25 daily fractions of 1.8 Gy treating 5 days a week for 5 weeks. Capecitabine 825 mg/m^2 BID orally (PO) was prescribed from the first day of radiation for 35 days (i.e. 7 days a week).

2.2.4. Toxicity assessment and DLT

Toxicities were graded according to the National Cancer Institute Common Terminology Criteria for Adverse Events version 3.0. Grade 3 and 4 toxicities were included in the toxicity analysis. Some specific toxicities were of particular interest and predefined in the protocol as DLTs. These included grade 3 diarrhoea that did not improve to grade 2 or less, within 24 h of commencing intensive antidiarrhoeal therapy; grade 4 diarrhoea; grade 3 or 4 fatigue; grade 3 or 4 febrile neutropenia; grade 3 or 4 infection; grade 4 thrombocytopenia associated with bleeding; grade 3 nausea/vomiting for at least 24 h despite full antiemetic treatment; grade 3 or 4 palmar-plantar erythrodysesthesia; grade 3 rash and a dose delay of more than 2 weeks because of drug-related toxicity. Abnormal

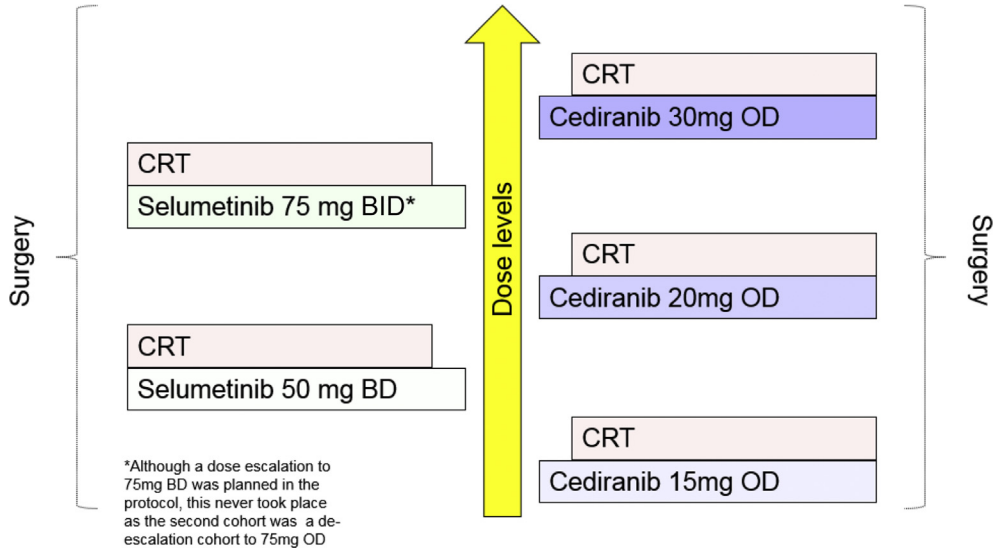

Fig. 1. DREAMtherapy trial schema showing the alternate use of cediranib and selumetinib. DREAMtherapy, Dual REctal Angiogenesis MEK inhibition radiotherapy, CRT, chemoradiotherapy; BID, twice a day; OD, once daily.

laboratory results were recorded for every patient with grade 3 and 4 values reported as toxicity.

2.2.5. Assessment of clinical efficacy

The patients were followed up for eight weeks after completion of treatment, at which point, a triphasic CT scan and pelvic MRI were performed with a view to tumour resection around 13 weeks after completion of treatment. For patients who achieved a complete radiological response, an examination under anaesthesia and biopsies were carried out. Complete clinical response (cCR) was defined as the absence of tumour radiologically, on examination and histologically. Patients with cCR were given the choice of intense surveillance or surgical resection. Pathological staging (Mandard) was determined in patients who underwent resection using a tumour regression grade (pTRG), where pTRG1 is a pCR [19].

Exploratory assessment of MRI tumour regression grade (mrTRG2) [20] was used as a potential surrogate marker of response. Pre- and post-treatment MRI scans were analysed, and five possible categories were defined: mrTRG1-no tumour signal; mrTRG2-predominant fibrosis (minimal tumour); mrTRG3-mixed fibrosis and tumour; mrTRG4-predominant tumour and mrTRG5-tumour (little fibrosis).

A pCR (pTRG1) and microfoci (pTRG2) have been compared, and both show similarly good outcome [21]. If these good prognostic groups are added to patients who have had a complete response without surgery (cCR), this has been termed as excellent clinical or pathological response (ECPR: cCR + pCR + microfoci), [7].

2.3. Translational research protocol

The patients provided blood samples for evaluation of circulating biomarker candidates and underwent dynamic contrast-enhanced MRI (DCE-MRI).

2.3.1. Plasma biomarkers

A panel of candidate biomarkers was evaluated in the cediranib arm. Two pre-treatment samples were taken: one at screening and the second on day 10, upon commencement of cediranib, to define the 95% confidence intervals (CIs) of reproducibility and thereby any therapeutic effect. Further samples were acquired at week 1 (day 1: before commencing CRT and after cediranib monotherapy), week 3 (midway through CRT) and week 5 (final week of CRT). A post-treatment sample was also taken at the end of the follow-up period (week 13) (see Fig. 2).

In cediranib-treated patients, we measured the plasma concentrations of a number of angiogenesis-associated candidate biomarkers, angiopoietin 1 and 2 (Ang1 and Ang2), basic fibroblast growth factor, hepatocyte growth factor, platelet derived growth factor B, VEGFs (VEGF-A, VEGF-C and VEGF-D), VEGF receptors 1 and 2 (VEGFR1 and VEGFR2), granulocyte colony-stimulating factor, keratinocyte growth factor, placental growth factor (PlGF), Tie2, stromal cell–derived factor 1β (SDF-1β), as well as tumour necrosis factor alpha (TNFα), interleukins 1b, 2, 5, 6 and 8 (IL-1b, IL-2, IL-5, IL-6 and IL-8), osteopontin, E-Selectin and vascular cell adhesion molecule 1 (VCAM1), using custom-made multiplexed enzyme-linked immunosorbent assays (ELISAs; Aushon Biosystems, Billerica, MA, USA). Endoglin (CD105) was analysed using individual ELISA kits (R&D Systems, Abingdon, UK). Cytokeratin 18 levels were measured as a surrogate of epithelial cell death using ELISA assays (PEVIVA [VL VBio], Bromma, Sweden). All of these assays were performed according to the manufacturer's instructions to Good Clinical Practice [22] in Clinical and Experimental Pharmacology (CRUK Manchester Institute). Blood samples were also collected from selumetinib-treated patients but were not analysed as only a small number of patients were recruited, and it was not possible to define a MTD.

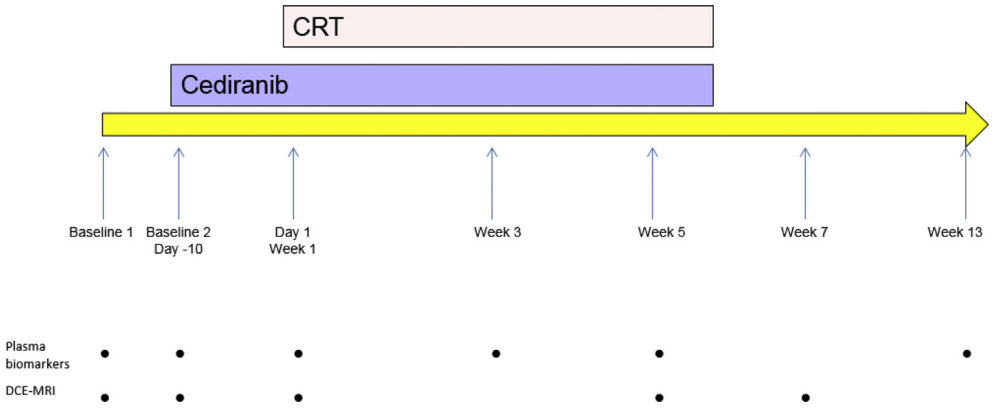

Fig. 2. DREAMtherapy schedule for translational protocol. DREAMtherapy, Dual REctal Angiogenesis MEK inhibition radiotherapy; CRT, chemoradiotherapy; DCE-MRI, dynamic contrast-enhanced magnetic resonance imaging.

2.3.2. Translational MRI method

DCE-MRI and diffusion-weighted imaging (DWI) were performed on a Philips 1.5 T Achieva scanner (Philips, Eindhoven, Netherlands). The scan protocol consisted of high-resolution T2-weighted and T1-weighted acquisitions followed by DWI (b values: 0, 150, 500 and 800). Next, native T1 estimation was performed using 3D spoiled volumetric grading echo acquisitions with variable flip angles (2°, 10°, 20° and 30°). Finally, a dynamic time series was acquired using the 30° flip angle acquisition with a temporal resolution of 4 s over a period of approximately 7 min [23]. Gadoterate meglumine (Dotarem; Guerbet S.A.) was administered as a single-dose bolus injection (0.1 mmol/kg body weight) after 10 dynamic acquisitions. Finally, a post-contrast T1-weighted sequence was acquired.

Regions of interest (ROIs) were outlined on the T2-weighted images using Osirix software (Pixmeo, Bernex, Switzerland) [24]. These areas were delineated by two investigators on all patients; one of them is an experienced radiologist with a specialist interest in gastrointestinal malignancies. The scans were also referenced to the diagnostic high-resolution standard-of-care MRI to delineate tumour accurately. Voxels within the bowel lumen were not included in the tumour ROI.

Imaging was performed pre-treatment, at week 1 day 1 of CRT and two weeks after completion of treatment (week 7). Two pre-treatment evaluations (separated by at least 2 days) were performed in the two weeks before administration of the first dose of cediranib to establish intrapatient variation in intratumour vascular parameters [23,25]. Based on the initial ROI delineated of the tumour, apparent diffusion coefficient (ADC) was derived from DWI using a monoexponential fit. Those voxels with zero ADC were excluded as these represent fitting errors. Kinetic analysis of the DCE-MRI data was carried out in MiStar (MiStar; Apollo Medical Imaging, Melbourne, Australia) using the modified Tofts model.

2.3.3. Statistical methods

To assess whether circulating and/or imaging biomarkers changed during treatment, a paired t-test was conducted with the corresponding *p*-value reported. To assess the correlation between pre-treatment biomarker values and ECPR, a logistic regression analysis was performed, and a *p*-value from the likelihood-ratio test was reported. In addition, box plots showing the distribution of pre-treatment biomarker values between the two types of response (ECPR vs non-ECPR) and receiver operating characteristic (ROC) area under the curve (AUC) with 95% CIs, and a threshold which maximises sensitivity/specificity was also reported. All plots and analyses were performed using R version 3.1.1.

Table 1
Summary of patient characteristics for the three cediranib cohorts.

Characteristic	Cohort 1 cediranib 15 mg/OD	Cohort 2 cediranib 20 mg/OD	Cohort 3 cediranib 30 mg/OD	Total number (%)
N	6	6	6	18
Sex				
Male	6	5	4	15 (83)
Female	0	1	2	3 (17)
Age				
Median	48	63	54	55
Range	42–58	56–70	42–70	42–70
Clinical staging				
T2	1[a]	1[a]	0	2 (11)
T3	5	5	4	14 (78)
T4	0	0	2	2 (11)
N0	2	0	1	3 (17)
N1	2	3	4	9 (50)
N2	2	3	1	6 (33)
Performance status				
0	6	5	5	16 (89)
1	0	1	1	2 (11)
Mean distance to anal verge (mm)	74.1	73.8	54.5	67.5
Mesorectal edge on MRI				
Not involved[b]	1	1	1	4 (22)
Potentially involved	1	0	1	1 (6)
Involved, not breached	4	4	4	12 (66)
Breached	0	1	0	1 (6)

MRI, magnetic resonance imaging; CRM, circumferential resection margin; OD, once a day.

[a] Both T2 tumours were considered to have threatened CRMs because of close proximity of the tumour to the seminal vesicles and the presence of a posterior node within 1 mm of the CRM, respectively.

[b] Tumours within 5 cm from the anal margin. For one of these patients, the tumour was thought to be within 5 cm from the anal verge (clinical examination) at the time of trial entry. Retrospective central review of MRI images located the tumour at 6.5 cm from the anal verge.

3. Cediranib cohort results

3.1. Patient characteristics

The demographic characteristics of the 18 patients are summarised in Table 1. One patient, who had histologically proven adenocarcinoma upon recruitment, received cediranib 15 mg OD but at resection was found to have a tumour with neuroendocrine differentiation (NET). All patients were evaluable for toxicity. The patient with a NET was not included in the response analysis.

3.2. Dose escalations, safety, toxicity and MTD

The patients were recruited to the three dose levels of cediranib, and the associated toxicities are shown in Table 2. Initially, 3 patients were recruited to the first and second cohorts, where no DLTs were reported. The third cohort was expanded to include six patients as one patient experienced dose-limiting grade 3 fatigue and diarrhoea. An additional DLT, grade 3 palmar-plantar erythema, was reported in the expanded cohort at this dose level, and 20 mg OD was defined as the MTD and the dose suitable for phase II evaluation. In view of the clinical activity seen in the 15-mg OD and 20-mg OD cohorts, ethical permission was sought to extend the recruitment within these cohorts to a total of six patients. Therefore, 18 patients were recruited into this arm of the trial (6 patients in each cohort).

All patients included in the study completed the planned radiotherapy treatment with the exception of one in the 30-mg OD cohort who missed two doses because of grade 3 dose-limiting lethargy and diarrhoea. In this patient, capecitabine was discontinued at week 4 of CRT. In the other patient, who experienced dose-limiting grade 3 palmar-plantar erythema (PPE), capecitabine was reduced by 25% during week 4 of the treatment and discontinued at week 5. An additional patient missed 5 days of capecitabine because of grade 3 neutropenia. Capecitabine was restarted with a 25% dose reduction after this episode. The remaining 15 patients (83%) had the capecitabine treatment in full. No delays in planned surgery occurred in any cohorts, and no significant postoperative complications were reported. Four patients had minimal postoperative wound healing difficulties, two of them taking more than 3 months to heal. However, no patients required further surgery.

The acute toxicity associated with the addition of cediranib to CRT appeared to be manageable in most cases. The expected toxicity associated with CRT was not exacerbated with incidence and was similar to that traditionally seen in treatment with CRT alone (Table 2). The toxicity directly attributable to cediranib was more prevalent in the 30-mg OD cohort, but no patients developed a fistula or bowel perforation. Based on the toxicity data, the recommended phase II dose of cediranib, in combination with CRT, is 20 mg OD.

3.3. Clinical activity

Of the five patients who were evaluable for response and treated with cediranib 15 mg OD, one had a cCR

Table 2
Recorded adverse events (AEs) for patients treated within the cediranib cohorts.

Adverse event	Cediranib 15 mg OD		Cediranib 20 mg OD		Cediranib 30 mg OD	
	Grade I/II	Grade III/IV	Grade I/II	Grade III/IV	Grade I/II	Grade III/IV
	N (%)	N (%)	N (%)	N (%)	N (%)	N (%)
Abdominal pain	2 (33)	–	1 (17)	–	2 (33)	–
Back pain	3 (33)	–	–	–	–	–
Constipation	–	–	2 (33)	–	3 (50)	–
Cystitis	4 (67)	–	3 (50)	–	–	–
Diarrhoea	2 (33)	1 (17)	5 (83)	–	4 (67)	1 (17)
Fatigue	2 (33)	–	4 (67)	–	5 (83)	1 (17)
Headache	2 (33)	–	1 (17)	–	3 (50)	–
Hoarse voice	–	–	1 (17)	–	4 (67)	–
Hyperbilirubinaemia	–	–	–	–	2 (33)	–
Hypertension	2 (33)	–	2 (33)	–	3 (50)	–
Mucositis	–	–	1 (17)	–	1 (17)	–
Nausea	2 (33)	–	1 (17)	–	1 (17)	–
Neutropenia						1 (17)
Palmar-plantar erythema	5 (83)	–	2 (33)	–	2 (33)	1 (17)
Perianal soreness	4 (67)	–	2 (33)	2 (33)	2 (33)	–
PR bleeding	2 (33)	–	2 (33)	–	–	–
Proteinuria	–	–	1 (17)	–	–	–
Rash	1 (17)	–	1 (17)	–	1 (17)	–
Rectal pain	1 (17)	–	1 (17)	–	1 (17)	–
Tenesmus	–	–	1 (17)	–	2 (17)	–

AEs reported in this table are those that were recorded in more than one patient. Full toxicity data are given in the Supplementary Information.
OD, once a day; PR, Per rectum.

Table 3
Summary of clinical efficacy in cediranib cohorts.

Cohort	Excellent clinical or pathological response (ECPR)			Non-ECPR (all responded but not an ECPR)	Other
	cCR (+mrTRG 1) (at 6 months)	pCR (pTRG1)	Residual microfoci (pTRG2)		
CRT + cediranib 15 mg/OD	1	2	1	1	1 patient was excluded from analysis (resection specimen was a NET)
CRT + cediranib 20 mg/OD	2	0	1	3	
CRT + cediranib 30 mg/OD	2	0	0	4	
Total	**5 (29%)**	**2 (12%)**	**2 (12%)**	**8**	1
Total ECPR	**9 patients (53%)**				
	7/9 patients had mrTRG1 or mrTRG2.			1/8 patients had mrTRG2 (none had mrTRG1).	

cCR, complete clinical response; pCR, pathological complete response; CRT, chemoradiotherapy; NET, tumour with neuroendocrine differentiation; mrTRG, magnetic resonance imaging tumour regression grade; OD, once a day.
Patients are classified as per either the radiological or pathological tumour regression grade (TRG).

(mrTRG1) [20] and two attained a pCR (pTRG1) [19], while two other patients had excellent radiological responses. One of the patients with excellent radiological response had only one focus of microscopic involvement on the resection specimen (pTRG2) (Table 3).

In the 20-mg OD cohort (n = 6), two had a cCR (mrTRG1), one had only microfoci on the resection specimen (pTRG2) and the other three patients had a good radiological response. In the final cohort (30 mg OD), two patients had a cCR (mrTRG1), and the other four patients responded radiologically. All patients underwent R0 complete resections.

If the patients with mrTRG1, pTRG1 and pTRG2 were defined as achieving an ECPR, nine patients (53%) in the cediranib arm would have been included. Seven of 17 patients achieved a clinical or pCR (41%).

In terms of MRI evaluation after CRT, 7 of 9 patients who had an ECPR attained mTRG1 or mTRG2. Only one patient of the 8 who did not have an ECPR had mTRG2. No mTRG1 were seen in patients who did not achieve an ECPR.

Five patients who had a cCR on MRI opted for careful clinical follow-up and avoided surgery. Three of these developed local recurrences at 9, 10 and 53 months, respectively, and all subsequently underwent R0 resections. Two other patients sustained cCR and remained disease free 70 and 80 months after radiotherapy. As of December 2017, there had not been any more local recurrences.

3.4. Translational analyses

The pre-treatment plasma concentrations of TNFα, PlGF, VEGF-D and IL-5 were lower in patients who attained an ECPR ($p < 0.001$, Fig. 3, Table 4). The difference of baseline TNFα values between ECPR and non-ECPR was found to be statistically significant with lower levels seen in the ECPR cohort. The ROC AUC for TNFα as a predictive biomarker was 0.89

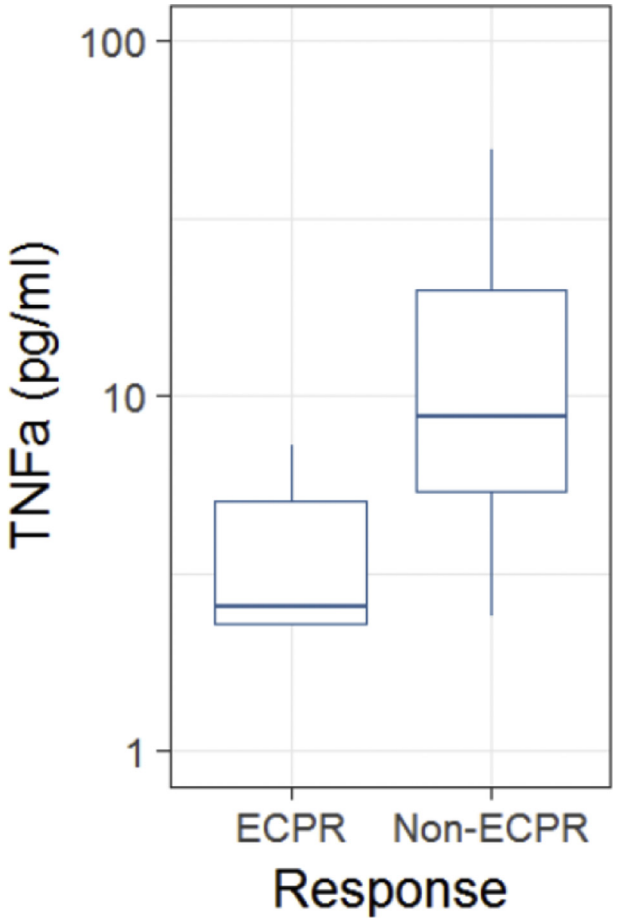

Fig. 3. Box plots showing the distribution of TNFα for patients with excellent clinicopathological response (ECPR) and non-ECPR. Circulating levels of TNFα were significantly different between the ECPR and non-ECPR groups ($p < 0.001$). The horizontal line within the box corresponds to the median, the bottom and top of the box are the 25th and 75th percentiles, respectively, and the whiskers denote the lowest and highest datum within 1.5 times the interquartile range. TNFα, tumour necrosis factor alpha.

Table 4
Summary statistic for selected baseline biomarkers.

Baseline biomarker	All patients Median (range)	ECPR Median (range)	Non-ECPR Median (range)
TNFα (pg/ml)	2.30 (5.05–1022.10)	2.55 (2.30–7.25)	14.40 (2.40–1022.10)
PlGF (pg/ml)	20.95 (2.00–206.10)	7.60 (2.00–185.60)	52.58 (7.40–206.10)
VEGF-D (pg/ml)	242.75 (19.50–20000.00)	169.83 (19.50–2906.85	557.73 (158.60–20000.00)
IL-5 (pg/ml)	1.50 (0.40–108.75)	0.55 (0.40–10.10)	6.05 (0.45–108.75)

TNFα = tumour necrosis factor alpha; PlGF = placental growth factor; VEGF-D = vascular endothelial growth factor D; IL-5 = interleukin 5.

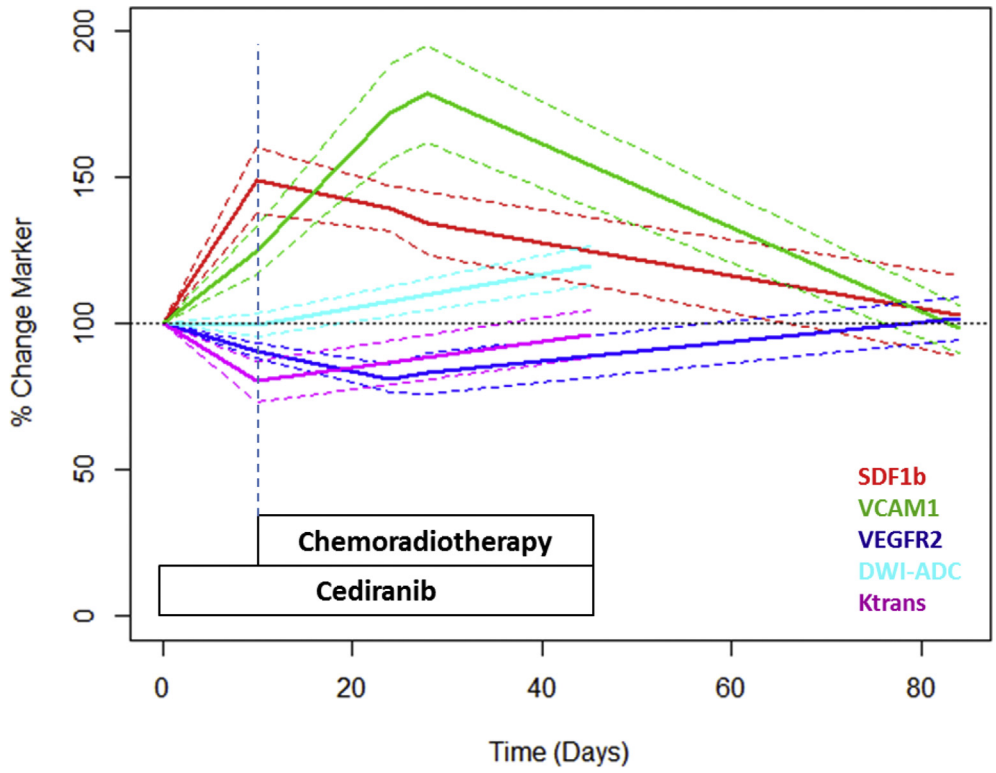

Fig. 4. Plot showing the mean change (solid) and standard error (dashed) for the time series of five markers of interest. SDF-1β, stromal cell–derived factor 1β; VCAM1, vascular cell adhesion molecule 1; VEGFR2, vascular endothelial growth factor receptor 2; ADC; apparent diffusion coefficient. DWI, diffusion-weighted imaging.

(95% CI: 0.7–1) (Supplementary Fig. S1). The optimal threshold of TNFα, which maximised sensitivity/specificity, was 5.3 pg/ml.

Fig. 4 shows the temporal changes in circulating and imaging biomarkers of interest over time. The maximum changes in SDF-1β and K^{trans} were observed during the administration of the single agent cediranib ($p = 0.002$ and $p = 0.061$ respectively). Maximum changes in VEGFR2, VCAM1 and DWI-MRI occurred during the combination treatment period such that in the nadir, VEGFR2 concentrations occurred at day 24 ($p = 0.005$), VCAM1 peaked at day 28 ($p < 0.001$) and an increase in ADC was seen on day 45 ($p = 0.015$). All three circulating biomarkers, SDF-1β, VEGFR2 and VCAM1, returned to the pre-treatment values by week 12 ($p = 0.703$, $p = 0.967$ and $p = 0.710$, respectively).

4. Selumetinib cohort results

4.1. Patient characteristics

Twelve patients were recruited to the selumetinib cohorts (Table 5). All had LARC including T3 or T4 tumours with nodal involvement.

4.2. Dose escalations, safety, toxicity and MTD

The protocol defined two cohorts for selumetinib: 50 mg BID and 75 mg BID. There was also a defined 'de-escalation' cohort (selumetinib 75 mg OD) in the event of a DLT being encountered in the initial cohort.

Eight patients were recruited to the selumetinib 50-mg BID cohort (see Table 5). Two of the initial 3

Table 5
Patient characteristics for the selumetinib cohorts.

Characteristic	Cohort 1 selumetinib 50 mg/BID	Cohort 0 selumetinib 75 mg/OD	Total number (%)
N	8	4	12
Sex			
Male	4	3	7 (58%)
Female	4	1	5 (42%)
Age			
Median	60	61	60
Range	(47–73)	(54–66)	(47–73)
Clinical staging			
T2[a]	1	0	1 (8%)
T3	7	0	7 (58%)
T4	0	4 (100%)	4 (34%)
N0	0	0	
N1	5	2 (50%)	7 (58%)
N2	3	2 (50%)	5 (42%)
Performance status			
0	7 (87%)	4 (100%)	11 (92%)
1	1 (13%)		1 (8%)

CRM, circumferential resection margin; BID, twice daily; OD, once a day.
[a] T2 tumour considered to have threatened CRM because of a node within 1 mm of the CRM.

Table 6
Summary of adverse events (AEs) seen in the selumetinib cohorts.

Adverse event	Selumetinib 50 mg BID		Selumetinib 75 mg OD	
	Grade I/II N (%)	Grade III/IV N (%)	Grade I/II N (%)	Grade III/IV N (%)
Abdominal pain	2 (25)	–	–	1 (25)
Anorexia	2 (25)	–	–	–
Back pain	2 (25)	–	–	–
Cheilitis	3 (38)	–	–	–
Constipation	5 (63)	–	–	–
Cystitis	3 (38)	–	1 (25)	1 (25)
Diarrhoea	7 (86)	1 (13)	2 (50)	2 (50)
Dizziness	2 (25)	1 (13)	–	–
Dyspepsia	1 (13)	–	1 (25)	–
Epiphora	1 (13)	–	1 (25)	–
Facial flushing	1 (13)	–	1 (25)	–
Fatigue	5 (63)	2 (25)	2 (50)	1 (25)
Mucositis	2 (25)	–	–	–
Nausea	5 (63)	–	2 (50)	–
Palmar-plantar erythema	4 (50)	–	1 (25)	–
Perianal soreness	4 (50)	–	2 (50)	–
Peripheral oedema	2 (25)	–	–	–
PR bleeding	1 (13)	1 (13)	–	–
Raised transaminases	1 (13)	3 (38)	–	1 (25)
Rash	7 (86)	–	2 (50)	–
Rectal pain	2 (25)	–	–	–
Tenesmus	2 (25)	–	–	–
Vomiting	2 (25)	–	–	1 (25)
Xerostomia	2 (25)	–	–	–

Only AEs recorded in more than one patient are included in this table. Full toxicity data are available in the Supplementary Information. BID, twice daily; OD, once a day; PR, Per rectum.

patients in the cohort were replaced as they experienced problems unrelated to the trial conduct early on the study (uncontrolled diabetes mellitus and rectal bleeding) but were considered evaluable for toxicity and response. One DLT was registered in this initial group of five patients (grade 3 diarrhoea), and thus, the cohort was expanded by further three patients so that 8 patients were treated in this cohort (see Table 6). A further DLT (grade 3 fatigue) was reported, and because two DLTs were reported in this cohort, the de-escalation cohort (75 mg OD) was opened. All patients in the 50-mg BID cohort received the planned radiotherapy. However, dose alterations of capecitabine and selumetinib were recorded in all patients apart from one.

Table 7
Summary of clinical efficacy in the selumetinib cohorts.

Cohort	6244 efficacy results		
	cCR (+mrTRG 1)	Responded and resected	No surgery due to MCRC
CRT+6244 Cohort 1 (50 mg BID)	0	6 (one patient had pTRG2)	2 (both never had LR until death)
CRT+6244 Cohort 1 (75 mg OD)	1	2	1 (tumour seen radiologically)
Total	**1 (8%)**	**8 (67%)**	**3 (25%)**

ECPR, excellent clinicopathological response; cCR, complete clinical response; pCR, pathological complete response; TRG, tumour regression grade; mrTRG, magnetic resonance imaging tumour regression grade. BID, twice daily; MCRC, metastatic colorectal cancer; LR, local recurrence; OD, once a day.
ECPR (cCR + pCR + pTRG2) = 2/12 = 17%.

In the de-escalation cohort, one of three patients experienced grade 3, dose-limiting diarrhoea. This cohort was therefore expanded, but the fourth patient also developed dose-limiting grade 3 diarrhoea, leading to closure of this cohort after entering a total of 4 patients. Three of 4 patients (75%) in the de-escalation cohort required dose modification to manage toxicity associated with either selumetinib or capecitabine or both.

4.3. Clinical activity

Of twelve patients, only two could be categorised as having an ECPR (17%) (Table 7). All eight patients were evaluable for response in the selumetinib 50-mg BID cohort. Six of these patients attained a response to treatment, and all proceeded to have R0 resections (one pTRG2). The remaining two patients, despite good response within the primary, developed progressive disease with widespread lymphadenopathy and liver metastases, respectively. In the 75-mg OD cohort, three of four patients responded to treatment. One patient had a cCR and was disease free 53 months after radiotherapy. Two patients responded well and proceeded to have a R0 resection. The fourth patient in this cohort was diagnosed with liver metastases upon completion of his neoadjuvant treatment.

5. Discussion

Conventional fluoropyrimidine-based CRT is routinely used, but only about 15% of patients achieve a pCR [1,2]. There is therefore a pressing need for improvement of this therapy to increase its effectiveness while maintaining organ function and quality of life. The potential novel combination of a traditional CRT backbone with a targeted agent opens a new range of therapeutic interventions in this cohort. There is also a need for novel trial strategies to evaluate these agents rapidly and in a cost-effective way.

In the DREAMtherapy (Dual REctal Angiogenesis MEK inhibition radiotherapy) trial, we have rapidly evaluated two novel drug–CRT combination regimens separately but within a single entwined study. In this alternating approach, when one cohort treated with a VEGF inhibitor (VEGFi)–chemoradiation combination was recovering and being assessed for toxicity, a second cohort was treated with a MEK inhibitor (MEKi)–chemoradiation regimen. While this cohort of patients was recovering, the second VEGFi cohort was treated at the second dose level and so on throughout the trial. At no point did the dual design represent a risk to the quality of the data, or indeed patient safety, as the two drugs were evaluated as separate entities within a single protocol. Moreover, although the trial was not randomised, it became evident that the cediranib arm efficacy and side–effect profile was very different from that encountered in the selumetinib cohort. We, therefore, propose this design as an alternative to reduce the inevitable natural gaps encountered when evaluating medium-term toxicity associated with radiotherapy. Another option would be to include a third 'calibration arm' to collect data to be compared in a non-randomised setting with the other two cohorts.

Seventeen patients received cediranib–chemoradiation; of whom, nine attained an ECPR (53%). Two had a pCR, and five, a cCR at 6 months, giving a cCR/pCR rate of 41%. This combination was therefore very effective in this small cohort of patients when compared with the 15% pCR rate that would normally be expected without the TKI. Three of the patients who had a cCR developed recurrent disease at 9, 10 and 53 months and underwent curative surgery as described by others [26]. This strategy is important as it is likely that clinicians or patients will choose not to have surgery in the event of a cCR. Thus, future trials will have to incorporate this end-point, reducing investigators' focus on pCR rate.

The administration of the CRT was not compromised by the addition of cediranib, which was crucial when considering that the treatment intent for this population is curative. All but one patient in this cohort completed all of the radiotherapy, and 15 patients (83%) had the capecitabine treatment in full. There were no delays in surgery, and no significant problems with wound healing or postoperative complications were reported. No reoperations were needed. If these data were compared

with trials that evaluated bevacizumab–CRT, then, it becomes clear that the use of a tyrosine kinase VEGFi is much better tolerated. Previous studies with bevacizumab were associated with significant toxicity, leading to early termination of some trials [13–15]. Potentially because of its long half-life, there were regular reports of wound healing difficulties and fistula formation in patients receiving bevacizumab–chemoradiation. The toxicity seen in this arm was largely because of the effects of CRT, and the toxicity attributed to cediranib was manageable. In the 30-mg OD cohort, dose-limiting grade 3 fatigue, diarrhoea and PPE were reported, leading to the 20-mg OD dose being defined as the MTD.

Circulating TNFα was identified as a potential predictive biomarker for the combination of cediranib with CRT. The patients who achieved an ECPR had a significantly lower pre-treatment plasma concentration of TNFα compared with those patients who had more modest responses. One potential explanation for this might be that inflammation and angiogenesis coexist [27] and that TNFα concentrations therefore indirectly reflect tumour angiogenesis and therefore potential response to VEGFi.

The combination of selumetinib with CRT proved to be more toxic than the cediranib combination. The most common toxicity in this cohort was diarrhoea, and the treatment was not tolerated even within the de-escalation cohort. Furthermore, the clinical efficacy appeared to be no better than previously published data [1,2]. In addition, although this trial was not randomised, it was difficult not to compare both drugs because the arms were entwined and included the same patient cohort treated at the same cancer centre. Accepting this limitation, the efficacy noted in the selumetinib cohort was considerably less than that seen in patients treated with cediranib–CRT. This is likely to be due to the increased toxicity leading to reduced systemic therapy because 10 of 12 patients (83%) had a reduction in either or both capecitabine and selumetinib. In addition, this might be explained by the inclusion of patients with KRAS-mutated tumours exclusively in the early stages of the study. The combination of this drug with docetaxel also did not meet the primary progression free survival (PFS) end-point in a KRAS mutation phase 3 non-small cell lung cancer (NSCLC) randomised control trial (RCT) [28]. Having said that, available data for the combination of sorafenib with CRT in KRAS-mutated tumours demonstrated clinical efficacy, suggesting that this lack of effect cannot be solely attributed to the choice of patients with KRAS-mutated tumours [29]. The benefit-risk for combination of selumetinib with CRT is not considered acceptable in the neoadjuvant setting, and this combination is not recommended for further study.

In conclusion, we have implemented a novel early-phase trial design, which allows efficient evaluation of two or more drugs given separately in combination with CRT. The combination of cediranib and CRT was well tolerated and showed promising clinical efficacy (cCR/pCR: 41%). The recommended dose of cediranib that will be taken into a multiarm, multistrategy platform trial is 20 mg OD. Circulating TNFα warrants further evaluation in prospective studies as a potential marker of response for the combination of cediranib and CRT.

Conflict of interest statement

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ejca.2019.04.029.

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