FOLFOXIRI in combination with panitumumab as first-line treatment in quadruple wild-type (KRAS, NRAS, HRAS, BRAF) metastatic colorectal cancer patients: a phase II trial by the Gruppo Oncologico Nord Ovest (GONO)

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Background: The FOLFOXIRI regimen developed by the Gruppo Oncologico Nord Ovest (GONO) demonstrated higher activity and efficacy compared with FOLFIRI in metastatic colorectal cancer (mCRC). Panitumumab is effective in some patients with KRAS codon 12–13 wild-type mCRC. KRAS codon 61, HRAS, NRAS, and BRAFV600E mutations might predict resistance to anti-epidermal growth factor receptor antibodies.

Patients and methods: We conducted a phase II study evaluating the combination of panitumumab (6 mg/kg on day 1) with a slightly modified GONO-FOLFOXIRI (irinotecan 150 mg/m², oxaliplatin 85 mg/m², and folinate 200 mg/m² on day 1, followed by fluorouracil 3000 mg/m² as a 48-h continuous infusion starting on day 1) repeated every 2 weeks as first-line treatment of wild-type KRAS, HRAS, NRAS (codon 12–13–61), and BRAF unresectable mCRC patients. Fluorouracil dose was reduced to 2400 mg/m² after two of the first three patients reported grade 3–4 diarrhoea (in one case with febrile neutropenia). Induction treatment was scheduled for a maximum of 12 cycles, followed by panitumumab ± fluorouracil/folinate maintenance until progression. Primary end point was overall response rate (ORR).

Results: Eighty-seven patients were screened and 37 were enrolled. Thirty-three patients achieved an objective response (ORR: 89%; 95% CI 75% to 96%). Sixteen patients (43%) underwent secondary surgery of metastases, and R0 resection was achieved in 13 cases (35%). At a median follow-up of 17.7 months, median progression-free survival was 11.3 months (95% CI 9.7–12.9 months). After amendment, most common grade 3–4 adverse events reported during induction treatment were neutropenia (48%; febrile neutropenia: 5%), diarrhoea (35%), asthenia (27%), stomatitis (14%), and skin toxic effect (14%). One treatment-related death was registered.

Conclusions: Adding panitumumab to FOLFOXIRI is feasible decreasing the dose of fluorouracil and irinotecan to reduce the risk of diarrhoea. Activity and secondary resectability of metastases among Ras–BRAF wild-type patients are promising.

Key words: BRAF, FOLFOXIRI, metastatic colorectal cancer, panitumumab, Ras

Introduction

The armamentarium against metastatic colorectal cancer (mCRC) has increased in the last decade, thanks to more effective chemotherapy and biologic agents such as bevacizumab, cetuximab, and panitumumab [1]. Recently, new molecules as albirecept and regorafenib further expanded the available treatment options in this setting [2]. Moreover, deeper insights into CRC biology [3] and the definitive recognition of the survival impact of surgical resection of metaseses [1] strengthened the need for a modulation of the intensity of first-line therapy in different patient subgroups.

Chemotherapy represents the backbone of treatment, and survival is linked with the administration of all the three active...
cytotoxic agents (fluorouracil/folinate, oxaliplatin, and irinotecan) in the course of the disease [4]. Moreover, chemotherapy activity is proved correlated with both efficacy [5] and secondary resectability of metastases in initially unreactable disease [6]. Moving from these data, the Gruppo Oncologico Nord Ovest (GONO) developed the FOLFOXIRI regimen (continuous infusion fluorouracil, folinate, oxaliplatin, and irinotecan), which demonstrated improved activity, secondary resection rate, and efficacy compared with FOLFIRI in a phase III trial [7, 8].

Cetuximab and panitumumab are effective in different lines of treatment in KRAS wild-type mCRC patients [9]. In particular, panitumumab combined with first-line FOLFOX4 reported higher overall response rate (ORR) and progression-free survival (PFS) over FOLFOX4 alone [10]. Phase III trials focused on mutations occurring in codon 12–13 of exon 2, which account for almost all KRAS mutations in CRC [3]; retrospective and post-hoc analyses suggest that rarer KRAS codon 61 mutations further improve patient selection [3, 11–13]. Moreover, BRAF V600E mutation (convincingly proved as a poor prognostic factor) [3] and mutations in other Ras family members (such as HRAS and NRAS) might identify an additional 10% of patients who are resistant to anti-epidermal growth factor receptor (EGFR) antibodies [3, 11–13].

Phase II trials evaluating triplet chemotherapy plus cetuximab reported interesting results in terms of activity, at the price of an increased rate of mucosal toxic effect (mainly diarrhoea) [14–17]. However, none of these trials prospectively enrolled patients on the basis of a Ras–BRAF wild-type status.

In the present trial, we evaluate the safety and the activity of panitumumab in combination with the GONO-FOLFOXIRI regimen in a 'quadruple wild-type' (KRAS, HRAS, NRAS, and BRAF) mCRC population.

**methods**

**patients**

Trial eligibility criteria were: histologically confirmed colorectal adenocarcinoma; KRAS, NRAS, and HRAS codon 12–13–61 and BRAF codon 600 wild-type status of primary CRC or metastasis; unreactable and measurable metastatic disease according to Response Evaluation Criteria in Solid Tumors (RECIST) version 1.1; age 18–75 years; Eastern Cooperative Oncology Group (ECOG) performance status (PS) 0–2 if age ≤70 years, or 0 if age 71–75 years; previous adjuvant chemotherapy completed more than 12 months before first relapse; no previous palliative chemotherapy for mCRC; no major surgical procedure within 28 days and no radiotherapy within 42 days before enrolment; adequate bone marrow, hepatic, and renal function; no central nervous system metastases, symptomatic cardiac disease, myocardial infarction, cerebrovascular accidents, or thromboembolic or haemorrhagic events in the past 6 months; no uncontrolled arrhythmia, active infections, inflammatory bowel disease, history of interstitial lung disease, or any other medical conditions clearly contraindicating the delivery of any chemotherapy.

The study was conducted in accordance to Helsinki declaration and Good Clinical Practice guidelines. Approval was gained from the institutional review board of all participating Institutions, and each patient gave written informed consent before enrolment.

**Ras and BRAF mutational analyses**

Mutational analyses carried out on formalin-fixed paraffin-embedded tumour samples were centralized at the Unit of Pathology, Department of Surgery, University of Pisa. Details about the applied methods are available with supplementary Appendix SA.1, available at Annals of Oncology online.

**procedures**

Treatment consisted of intravenous therapy with panitumumab 6 mg/kg over 1 h, followed by irinotecan 150 mg/m² over 1 h, followed by oxaliplatin 85 mg/m² and folinate 200 mg/m² concomitantly over 2 h through a Y-connector, on day 1, and followed by fluorouracil 3000 mg/m² as a 48-h continuous infusion starting on day 1. After amendment, fluorouracil dose was reduced to 2400 mg/m². Induction treatment was administered every 2 weeks for a maximum of 12 cycles, or fewer cycles in case of disease progression, unacceptable toxic effect, or withdrawal of consent. Resectability of metastases was assessed every 2 months and strongly recommended when feasible. Unreactability was established according to the OncoSurge criteria [18], by a multidisciplinary team comprising liver surgeons and medical oncologists from participating centres. After metastasectomy, induction treatment was resumed until the completion of the 12 established cycles. After 12 cycles, patients continued on panitumumab ± fluorouracil/folinate at the same schedule, administered until progressive disease, unacceptable toxic effects, or patient refusal.

**design and statistical analysis**

This is an open-label, single-arm, phase II study. Primary end point was ORR by RECIST 1.1 in the intention-to-treat population. Secondary end points were PFS, R0 surgery on metastases, overall survival (OS), safety, and analyses of surrogate markers of activity.

The assessment of response was based on investigator-reported measurements that were confirmed by a central review, carried out by the coordinating centre. PFS was measured from the day of treatment start until the date of disease progression or death for any cause. OS was measured from the day of treatment start until the date of death for any cause, censoring patients who had not died at the last date known to be alive. The probability of progression and survival was calculated using the Kaplan–Meier method.

Toxicity was evaluated every 2 weeks according to National Cancer Institute—Common Toxic effect Criteria (version 3.0). The enrolling centre was responsible for gathering data for each patient.

The ORR reported with FOLFOXIRI is around 60%: an ORR of ≥80% should be considered promising. According to a Simon’s minimax two-stage design, assuming an alpha and beta errors of 0.05 and 0.20, respectively, and selecting p0 (ORR in null hypothesis) = 0.60 and p1 (ORR in alternative hypothesis) = 0.80, in the first stage of the study we needed to observe at least 9 responses among 13 patients. In the second stage, 26 responses over 35 patients would have been necessary to meet the primary end point. Statistical analyses were done using GraphPad Prism and SPSS package.

This study is registered with ClinicalTrials.gov, number NCT01358812.

**results**

**patients**

From 25 March 2010, to 15 October 2011, 87 patients were screened at five Italian Institutions. Thirty-seven (43%) patients were enrolled (Table 1). Figure 1 shows the trial profile. As expected, mutations in the analysed genes were mutually exclusive.
Table 1. Patient demographic and clinical characteristics

<table>
<thead>
<tr>
<th></th>
<th>Enrolled patients (n = 37)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age (years)</strong></td>
<td></td>
</tr>
<tr>
<td>Median</td>
<td>63</td>
</tr>
<tr>
<td>Range</td>
<td>33–72</td>
</tr>
<tr>
<td><strong>Gender</strong></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>21 (57%)</td>
</tr>
<tr>
<td>ECOG performance status</td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>28 (76%)</td>
</tr>
<tr>
<td>1</td>
<td>9 (24%)</td>
</tr>
<tr>
<td><strong>Primary tumour</strong></td>
<td></td>
</tr>
<tr>
<td>Colon</td>
<td>26 (70%)</td>
</tr>
<tr>
<td>Rectum</td>
<td>11 (30%)</td>
</tr>
<tr>
<td><strong>Surgery for primary tumour</strong></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>23 (62%)</td>
</tr>
<tr>
<td>No</td>
<td>14 (38%)</td>
</tr>
<tr>
<td><strong>Previous adjuvant chemotherapy</strong></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>7 (19%)</td>
</tr>
<tr>
<td>No</td>
<td>31 (84%)</td>
</tr>
<tr>
<td><strong>Previous adjuvant oxaliplatin</strong></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>6 (16%)</td>
</tr>
<tr>
<td>No</td>
<td>31 (84%)</td>
</tr>
<tr>
<td><strong>Disease sites</strong></td>
<td></td>
</tr>
<tr>
<td>Single</td>
<td>20 (54%)</td>
</tr>
<tr>
<td>Multiple</td>
<td>17 (46%)</td>
</tr>
<tr>
<td><strong>Synchronous metastases</strong></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>25 (68%)</td>
</tr>
<tr>
<td>No</td>
<td>12 (32%)</td>
</tr>
<tr>
<td><strong>Liver-only metastases</strong></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>12 (32%)</td>
</tr>
<tr>
<td>No</td>
<td>25 (68%)</td>
</tr>
</tbody>
</table>

Data are number (%), unless otherwise indicated.

ECOG, Eastern Cooperative Oncology Group.

efficacy

One (3%) complete response (CR) and 32 (86%) partial responses (PRs) were observed, for an ORR of 89% (95% CI 75% to 96%) at central radiologic review: 20 (54%) patients had their tumour shrunk more than 50% of the original measurement (supplementary Figure S1, available at Annals of Oncology online). Three (8%) patients achieved disease stabilization and 1 (3%) progressed at first evaluation. All the 12 patients with liver-limited disease obtained a PR (ORR: 100%; 95% CI 74% to 100%) (supplementary Table S1, available at Annals of Oncology online). Median time to response was 61 days (range 47–176 days).

Secondary surgery (in some cases with intra-operative thermoablation) on metastases was attempted with a curative intent in 16 (43%) patients: 14 patients were resected at the time of best response, and 2 patients after progression (due to radiological CR or delayed surgery for logistic reasons). A median of 10.5 pre-operative cycles (range 5–15) was administered. The median time between the last treatment administration and surgery was 34 days (range 19–113 days).

R0 resection was achieved in 13 (35%) patients: 1 patient had R1 lymphadenectomy, and 2 did not complete the scheduled two-stage hepatectomy and were classified as R2 resections. Five (38% of the R0 population) patients received liver resection, 1 (8%) liver resection plus thermoablation, 2 (15%) primary tumour and liver resection, 3 (23%) primary tumour and liver resection plus thermoablation, 1 (8%) lymph-node resection, and 1 (8%) lymph-node and uterus resection.

Among patients with liver-only metastases, the R0 resection rate was 75% (nine patients). Pathological CR was noted in 3 (23%) of the R0-resected patients.

At the time of the analysis, all patients are off treatment except for three continuing maintenance therapy. After a median follow-up of 17.7 months, 28 (76%) patients have progressed and the median PFS was 11.3 months (range 1.8–20.0; 95% CI 9.7–12.9 months; Figure 2A). Only 9 (24%) patients have died and median OS is thus not yet reached (Figure 2B). Median PFS among patients with liver-limited disease was 14.2 months (range 3.5–20.0; 95% CI 7.5–20.9 months) (supplementary Table S1, available at Annals of Oncology online).

safety

With fluorouracil 3000 mg/m², two of the first three patients reported serious adverse events (SAEs) (grade 4 diarrhoea plus febrile neutropenia and grade 3 diarrhoea) leading to hospitalization and resolved without sequelae. After amendment, a total of 357 induction cycles with a median of 11 cycles (range 3–16) per patient were administered. The relative dose intensity was about 75% of that planned for chemotherapy (fluorouracil: 76%; oxaliplatin: 75%; irinotecan: 74%) and 81% for panitumumab. Treatment was delayed because of toxic effect in 34 (10%) cycles, 16 (4%) of which due to SAEs.

After amendment, the only grade 3–4 haematological toxic effect reported during induction treatment was neutropenia (n = 18 [48%]) (Table 2). Only 2 (5%) patients experienced febrile neutropenia. The use of granulocyte colony-stimulating factor as secondary prophylaxis was administered in 46 (13%) cycles. Grade 3–4 non-haematological adverse events reported in more than 10% of the patients were diarrhoea (n = 13 [35%]), asthenia (n = 10 [27%]), stomatitis (n = 5 [14%]), skin toxic effect (n = 5 [14%]), and nausea (n = 4 [11%]) (Table 2).

A total of 149 cycles of maintenance treatment with panitumumab (97%) and fluorouracil/folinate (34%) were administered: the only grade 3 adverse events observed were skin toxic effect (n = 5 [24%]), stomatitis, peripheral neurotoxicity, and neutropenia (1 [5%] patient each).

After amendment 19 SAEs in 13 patients occurred. Two SAEs (one judged related to study procedures) resulted in patient death. One patient experienced febrile neutropenia and sepsis from Klebsiella Pneumoniae after the third cycle: toxic effect rapidly improved by inpatient management, but he died 3 weeks after due to general conditions deterioration. Another patient underwent sudden cardiac death 9 days after R0 resection of the rectal primary tumour and liver metastases. No peri-operative complications had occurred. Patient history revealed coronary artery bypass graft (10 years before enrolment), type II diabetes mellitus, hypercholesterolaemia, and arterial hypertension. All the other SAEs required hospitalization but resolved without sequelae (supplementary Table S2, available at Annals of Oncology online).
discussion

The present trial shows that combining panitumumab with the GONO-FOLFOXIRI regimen is feasible with chemotherapy dose adjustments and results are promising among Ras–BRAF selected patients. Moving from the available safety data, we initially scheduled a slight reduction in the dose of fluorouracil and irinotecan compared with the standard GONO regimen. Subsequently, we further reduced the dose of fluorouracil, not to decrease substantially the dose intensity of irinotecan. The safety profile reported thereafter appears consistent with the available literature data for triplet chemotherapy with or without biologics: neutropenia, skin toxic effect, diarrhoea, and stomatitis were the most common adverse events. The rate of severe neutropenia (48%) is comparable to that reported with FOLFOXIRI alone [7] or combined with bevacizumab [19, 20], and few patients experienced febrile neutropenia (5%). Most relevant appears the impact of mucosal toxic effect, as reported also by other groups with different triplet chemotherapy schedules plus cetuximab [14–17]: in this regard, our schedule seems associated with a more favourable safety profile. However, considering the 35% rate of severe diarrhoea not acceptable in patients selected with regard to age and PS, we scheduled a reduction of irinotecan to 130 mg/m² for our ongoing trial with FOLFOXIRI plus cetuximab [21].

Incidence of Ras members (39%) and BRAF (5%) mutations are comparable to that observed in other first-line trials. Beyond KRAS codon 12–13 mutations, KRAS codon 61, BRAF V600E, HRAS, and NRAS mutations are the most promising
and widely studied biomarkers for anti-EGFR agents [13]. In this molecularly screened population, our approach achieved a promising ORR (89%), which is, to our knowledge, the highest reported with a triplet plus a biologic in the so-far largest trial among selected patients. This resulted into relevant secondary (43%) and R0 (35%) resection rates. Results appear even more promising among patients with liver-limited disease, making this strategy suitable for selected patients with potentially resectable liver-only metastases.

The phase III TRIBE trial has recently showed that FOLFOXIRI plus bevacizumab achieved superior ORR and PFS compared with FOLFIRI plus bevacizumab in molecularly unselected mCRC patients [20]. As a result of a less favourable non-haematological safety profile, in the present trial a higher percentage of patients (27% versus 4% in Masi et al. [19]) discontinued induction therapy due to toxic effect. It is arguable, however, that a particularly active combination may achieve rapid tumour response: this could be of interest in the case of symptomatic disease (to provide a quick palliative effect) or potentially resectable tumour (to reduce treatment-induced liver injury). Therefore, alternative strategies aiming to shorten the duration of intensive chemotherapy are needed. We are now conducting a randomized phase II trial evaluating single-agent cetuximab or bevacizumab as maintenance therapy after 4 months of FOLFOXIRI plus cetuximab [21].

The present trial has several limitations. Patients have been strictly selected from molecular and clinical perspectives: therefore, the reported results cannot be translated to the whole KRAS codon 12–13 wild-type population. Moreover, sample size is small, and selection bias may have contributed to the high ORR (e.g. by excluding poor-prognosis BRAF mutant patients) and secondary resection rate (e.g. by selecting upfront patients with potentially resectable disease). Finally, the investigational approach is not compared with a standard doublet-plus-biologic strategy: it could be suggested that the results of first-line therapy may be improved by improving molecular patient selection alone, rather than by intensifying chemotherapy.

To conclude, we demonstrate that the combination of panitumumab and FOLFOXIRI is safe with dose reductions of fluorouracil and irinotecan due to high rate of diarrhoea. In patients selected for a quadruple (KRAS, HRAS, NRAS, and BRAF) wild-type disease, the combination achieved interesting results in terms of activity and secondary metastasectomy. Future trials will evaluate a shorter duration of induction therapy and clarify the role of triplet compared with doublet regimens in molecularly screened mCRC patients.

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funding

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disclosure

AF is a consultant for Amgen, Merck, and Roche and has received honoraria and expenses for travel, accommodations, and meetings from Amgen, Merck, and Roche. All remaining authors have declared no conflicts of interest.

references