Primary tumor location and bevacizumab effectiveness in patients with metastatic colorectal cancer


Departments of 1Oncology, 2Medicine Herlev Hospital, Copenhagen University Hospital, Herlev, 3Statistics Bioinformatics and Registry, Danish Cancer Society, Copenhagen; 4Department of Oncology Roskilde Sygehus, Roskilde; 5Department of Oncology Righospital, Copenhagen University Hospital, Copenhagen, Denmark; 6Department of Oncology Västerås County Hospital, Västerås, Sweden; 7Department of Oncology Aalborg Sygehus, Aalborg; 8Department of Oncology and Palliation Hillerød Hospital, Hillerød; 9Department of Oncology Odense University Hospital, Odense; 10Department of Oncology and Hematology Næstved Sygehus, Næstved; 11Department of Oncology Herning Hospital, Herning; 12Department of Oncology Vejle Sygehus, Vejle; 13Department of Oncology Sydvestjysk Sygehus, Esbjerg, Denmark

Received 16 April 2013; revised 27 May 2013; accepted 28 May 2013

Background: There is an unmet need for predictive markers for the antiangiogenic agent bevacizumab in metastatic colorectal cancer (mCRC). We aimed to assess whether the location of the primary tumor is associated with bevacizumab effectiveness when combined with capcitabine and oxaliplatin (CAPEOX) in the first-line treatment of patients with mCRC.

Patients and methods: A cohort of 667 consecutive patients with mCRC from the general community treated from 2006 to 2011 with CAPEOX and bevacizumab as standard first-line therapy was compared with a cohort of 213 patients treated with CAPEOX from 2003 to 2006, before bevacizumab was approved. Main outcome measures were progression-free survival (PFS) and overall survival (OS). Differences in outcome were tested using Kaplan–Meier curves and log-rank tests, and multivariate analyses were carried out using Cox Proportional Hazards models.

Results: Patients treated with CAPEOX and bevacizumab with primary tumors originating in the sigmoid colon and rectum had a significantly better outcome than patients with primary tumors originating from the cecum to the descending colon, both for PFS (median PFS 9.3 versus 7.2 months; hazard ratio (HR) 0.68, 95% confidence interval (CI) 0.56–0.82) and for OS (median OS 23.5 versus 13.0 months; HR 0.47, 95% CI 0.38–0.57). This difference was confirmed in multivariate analyses after adjustment for other potentially prognostic factors. For patients treated with CAPEOX, there was no association between primary tumor location and outcome, neither in unadjusted nor adjusted analyses.

Conclusions: The addition of bevacizumab to CAPEOX in first-line treatment of patients with mCRC may primarily benefit patients with primary tumors originating in the rectum and sigmoid colon. This hypothesis needs to be validated in data from completed randomized trials.

ClinicalTrials.gov identification number: NCT00212615.

Key words: metastatic colorectal cancer, bevacizumab, chemotherapy, primary tumor, biomarker
Introduction

Colorectal cancer (CRC) is a leading cause of cancer-related mortality worldwide [1, 2]. Although still modest, the survival for patients with metastatic CRC (mCRC) has increased during the last two decades, partly as a result of the introduction of new effective chemotherapeutics and biologic agents such as bevacizumab (Avastin®, Roche), targeting the vascular endothelial growth factor (VEGF-A), and cetuximab and panitumumab targeting the epithelial growth factor receptor (EGFR) [3–8]. Lack of treatment efficacy remains a major problem in mCRC, partly because of a lack of predictive markers. Mutation in the Kirsten rat sarcoma (KRAS) oncogene is currently the best selection marker used for anti-EGFR therapy [9]. No markers are clinically available for selection of patients with mCRC to treatment with bevacizumab [10].

Tumors from the rectum and proximal and distal segments of the colon can be very different macroscopically, and they may also exhibit different genetic and epigenetic alterations [11]. The tissue expression of VEGF-A, the target of bevacizumab, has also been demonstrated to vary depending on the location of the primary tumor, with higher expression observed in tumors from the distal colon and rectum than in tumors from the proximal colon [12].

We investigated the impact of the primary tumor location on outcome in two large cohorts of patients with mCRC treated with first-line capcitabine and oxaliplatin with or without bevacizumab (CAPEOXBEV/CAPEOX) to determine whether the primary tumor location was associated with bevacizumab effectiveness.

data extraction and end points

Data were extracted from individual patient records and electronic databases at each hospital using a standardized case report form. Pathology data and survival status were extracted from national databases using the unique Civil Registration System number assigned to every Danish citizen. This allowed for complete information about all pathologic diagnoses and updated survival status for every patient. Primary tumor location was registered as (i) cecum and ascending colon, (ii) right flexure and transverse colon, (iii) left flexure and descending colon, (iv) sigmoid and rectosigmoid colon, or (v) rectum. Survival status was updated on 29 November 2012. Date of disease progression was defined as the date of an evaluation CT scan showing progression according to the RECIST 1.0 criteria or, in a few cases, by other clinical signs of progression, if a diagnosis of clinical progression was stated unequivocally in the patient record. The end point progression-free survival (PFS) was measured from the initiation of treatment to disease progression or death from any cause. If patients died without evidence of disease progression more than 3 months after the last evaluation scan, they were censored at last known date of nonprogression. Overall survival (OS) was measured from initiation of treatment to death from any cause.

statistical analysis

The primary tumor location variable was categorized according to the five groups used in the case report form. PFS and OS were initially analyzed using Kaplan–Meier curves, and differences between survival curves were tested using the log-rank test. Cox Proportional Hazards models were applied for estimating the effect of covariates on PFS and OS on the two cohorts in two separate analyses. Subsequently, we combined the two cohorts and tested the interaction between cohort and location by means of a likelihood ratio test. Student’s t-test was used for comparison of means and proportions and Pearson’s χ² test was used for comparison of distributions. The statistical software packages R [15] (www.r-project.org) and GraphPad Prism 5 (GraphPad Software, Inc.) were used for all analyses. In all tests, a significance level of 0.05 was used.

Ethics

The BETmiRC study has been approved by the regional scientific ethical committee (approval number H-1-2010-081).

results

The study population comprised 880 patients; 667 treated with CAPEOXBEV and 213 treated with CAPEOX (Table 1). The distribution of primary tumor locations was similar in the two cohorts, but the cohorts differed significantly regarding other characteristics. The CAPEOXBEV patients were older, and had a higher proportion of female gender, performance status 0, intact primary, and synchronous metastatic disease at baseline. The CAPEOXBEV patients on average received one more cycle of first-line chemotherapy, but less often received later-line irinotecan and cetuximab, most likely as a result of shorter follow-up in this cohort.

Patients treated with CAPEOXBEV clearly separated into two primary location groups with differing outcome (Figure 1). Patients with primary tumors in the sigmoid colon and rectum had a significantly longer PFS (median PFS 9.3 versus 7.2 months; hazard ratio (HR) 0.68, 95% confidence interval (CI) 0.56–0.82) and OS (median OS 23.5 versus 13.0 months; HR 0.47, 95% CI 0.38–0.57) than patients with primary tumors

methods

patients

The clinical database from the bevacizumab tissue microRNAs in colorectal cancer (BETmiRC) biomarker study was used for the analyses. This study retrospectively included patients with mCRC treated with first-line CAPEOXBEV from 10 departments of oncology in Denmark in the period 2006–2011. These 10 sites encompassed all of the departments in the country which identified CAPEOXBEV as one of their preferred first-line treatments for patients with mCRC. Inclusion criteria were biopsy-confirmed adenocarcinoma of the colon or rectum and first-line systemic treatment of metastatic disease with CAPEOXBEV. Exclusion criteria were other coexisting malignancy, endocrine histology, and CAPEOXBEV given explicitly as adjuvant treatment. Disease evaluation was done every three to four cycles using computed tomography (CT) scans and the Response Evaluation Criteria in Solid Tumors (RECIST) 1.0 criteria [13]. We also included a second cohort of patients treated with CAPEOX using the same inclusion and exclusion criteria, except that later-line bevacizumab was not allowed. These patients were included from Herlev University Hospital and from a randomized study carried out in this period [14]. This second cohort started CAPEOX between 2003 and 2006, before bevacizumab was adopted as standard first-line therapy in Denmark. Treatment was given as infusional oxaliplatin 130 mg/m² on day 1, capcitabine 2000 mg/m² daily day 1–14, ±infusional bevacizumab 7.5 mg/kg on day 1, repeated every 3 weeks. More than 98% of patients were Caucasian.
from the cecum to the descending colon. Removing cases where patients were treated in later lines with cetuximab or irinotecan did not alter the difference observed for OS (supplementary Figure S1, available at Annals of Oncology online).

In patients treated with CAPEOX, there was no evidence of difference in outcome for different primary tumor locations, with median PFS ranging from 6.5 to 7.8 months and median OS ranging from 14.1 to 18.4 months (Figure 1). There were no statistically significant differences in PFS or OS between trial and nontrial patients in this cohort (data not shown).

When dividing patients into colon and rectal cancer subgroups, a significant difference in OS favoring patients with rectal cancer was observed for patients treated with CAPEOXBEV (median OS 24.1 versus 16.6 months; HR 0.73, 95% CI 0.61–0.87), but not for patients treated with CAPEOX alone (median OS 14.1 versus 16.4 months; HR 1.02, 95% CI 0.77–1.35).

In multivariate analyses, after adjustment for potentially prognostic factors (age, sex, resected primary tumor, treating department, prior adjuvant chemotherapy, number of metastatic sites, performance status, white blood cell count, and alkaline phosphatase level), the statistically significant difference in outcome between location groups was maintained for patients treated with CAPEOXBEV, while no statistically significant differences were seen for patients treated with CAPEOX (Table 2). Removing cases with missing information about one or more of the potentially prognostic variables did not change the differences observed between location groups in the CAPEOXBEV cohort (supplementary Table S2, available at Annals of Oncology online).

When dividing all patients from both treatment cohorts by location of the primary tumor into the two above-mentioned groups (cecum to descending colon versus sigmoid colon and rectum), a significant interaction was observed between the effect of bevacizumab and location group for OS ($P = 0.004$) but not for PFS ($P = 0.15$).

**Discussion**

In this exploratory analysis, we have provided evidence for a possible interaction between location of the primary tumor and effectiveness of bevacizumab treatment in patients with mCRC. The data were collected from a cohort of Danish patients uniformly treated nationwide with the same standard treatment. When stratifying for precise location of the primary tumor,
patients treated with CAPEOXBEV clearly divided into two prognostic groups; a poor-prognosis group consisting of patients with primary tumors from the cecum to the descending colon and a good prognosis group consisting of patients with primary tumors from the sigmoid colon and rectum. This difference in outcome was maintained when adjusting for potentially prognostic factors in a multivariate analysis. For patients treated with CAPEOX without bevacizumab, there was no correlation between primary tumor location and outcome.

Bendardaf et al. [12] have shown that VEGF-A, the target of bevacizumab, is present in higher levels in left-sided colon and rectal cancers than in right-sided colon cancers. If cancers originating in the left colon and rectum are more VEGF dependent, this could explain the improved outcome with VEGF inhibition observed for these primary tumor locations. Right-sided colon cancers are more often hypermutated (microsatellite instability), hypermethylated (CpG island methylator phenotype), BRAF-mutated, and more often have BRAF-mutation-like gene expression [10, 11, 16]. These differences can harbor prognostic and predictive value, e.g. BRAF-mutation(-like) and anti-EGFR treatment, but they have generally not yet been investigated in relation to anti-VEGF treatment. It would have been relevant to correct for the influence of BRAF mutation in our study, but the BRAF-mutation status was only available for a few patients.

The observed differences in outcome were greater for OS than for PFS. This could be explained by the frequent use of bevacizumab beyond first line (25% of patients). In addition, the magnitude and severity of disease progression at the time of diagnosis of progression according to established criteria, in this case RECIST, can be quite variable and, therefore, time-to-progression end points are not always good proxies for survival [17]. As exclusion of patients treated with cetuximab or irinotecan did not significantly alter the results, differing efficacies of these agents are likely not the main explanation for the larger OS differences.

Data from prospective clinical trials relating precise primary tumor location to bevacizumab efficacy is currently not publicly available.

**Figure 1.** Kaplan–Meier plots for progression-free survival and overall survival stratified by location of primary tumor. CAPEOX, capecitabine + oxaliplatin; PFS, progression-free survival; OS, overall survival; CA, caecum and ascending colon; RT, right flexure and transverse colon; LD, left flexure and descending colon; SI, sigmoid colon; RE, rectum.
Likelihood ratio test for interaction between CAPEOX cohort (accordance with the mentioned prospective trial data. not when treated with chemotherapy alone, which is in cancer when treated with chemotherapy and bevacizumab, but significantly longer OS for patients with rectal versus colon cancer [4, 6, 21–24]. In addition, the possible difference in progression timing. Yet, since OS, which was not affected by this uncertainty, showed the same prognostic division as PFS, this is unlikely to have had a significant effect on the results. Excluding cases with missing data did also not alter the results significantly. The cohort treated with CAPEOX without bevacizumab was treated in another time period, just before bevacizumab was approved, and therefore could be less comparable to the CAPEOXBEV cohort. The two cohorts were also not balanced regarding baseline variables. However, we corrected for the impact of these variables in the multivariate analyses, both within the cohorts separately and in the combined cohort, to account for this. The chemotherapy-only cohort was smaller than the bevacizumab-treated cohort which, of course, weakens the evidence in the results obtained from this cohort.

On the other hand, the difference in outcome we observe seems unequivocal and the effect is large, which makes it less likely to be a chance finding. In addition, the possible difference in efficacy of bevacizumab for different primary tumor locations was supported by findings from published randomized and observational trials investigating the differential effects of chemotherapy with or without bevacizumab in rectal versus colon cancer [4–6, 18–24]. The addition of bevacizumab to CAPEOX in the first-line treatment of patients with mCRC may primarily benefit patients.

Table 2. Multivariate Cox proportional hazards models

<table>
<thead>
<tr>
<th>CAPEOXBEV cohort (N = 654)a</th>
<th>Progression-free survival (PFS)</th>
<th>Overall survival (OS)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Hazard ratiob (95% CI) P</td>
<td>Hazard ratiob (95% CI) P</td>
</tr>
<tr>
<td>Primary tumor location</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cecum and ascending colon</td>
<td>1 [reference] – 1 [reference] –</td>
<td></td>
</tr>
<tr>
<td>Right flexure and transverse colon</td>
<td>0.78 (0.55–1.12) 0.17</td>
<td>0.89 (0.64–1.24) 0.49</td>
</tr>
<tr>
<td>Left flexure and descending colon</td>
<td>0.99 (0.66–1.49) 0.96</td>
<td>0.97 (0.65–1.46) 0.89</td>
</tr>
<tr>
<td>Sigmoid colon</td>
<td>0.64 (0.50–0.83) 0.0006</td>
<td>0.52 (0.40–0.67) &lt;0.0001</td>
</tr>
<tr>
<td>Rectum</td>
<td>0.65 (0.51–0.84) 0.001</td>
<td>0.46 (0.35–0.59) &lt;0.0001</td>
</tr>
<tr>
<td>Cecum to descending colon</td>
<td>1 [reference] 1 [reference] –</td>
<td></td>
</tr>
<tr>
<td>Sigmoid colon and rectum</td>
<td>0.69 (0.57–0.84) 0.0002</td>
<td>0.51 (0.42–0.61) &lt;0.0001</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>CAPEOX cohort (N = 211)a</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Primary tumor location</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cecum and ascending colon</td>
<td>1 [reference] – 1 [reference] –</td>
<td></td>
</tr>
<tr>
<td>Right flexure and transverse colon</td>
<td>1.01 (0.51–2.01) 0.98</td>
<td>0.49 (0.25–0.99) 0.047</td>
</tr>
<tr>
<td>Left flexure and descending colon</td>
<td>0.89 (0.42–1.87) 0.75</td>
<td>0.89 (0.43–1.85) 0.76</td>
</tr>
<tr>
<td>Sigmoid colon</td>
<td>0.92 (0.57–1.50) 0.74</td>
<td>0.74 (0.46–1.20) 0.23</td>
</tr>
<tr>
<td>Rectum</td>
<td>0.85 (0.52–1.39) 0.52</td>
<td>0.97 (0.62–1.52) 0.89</td>
</tr>
<tr>
<td>Cecum to descending colon</td>
<td>1 [reference] 1 [reference] –</td>
<td></td>
</tr>
<tr>
<td>Sigmoid colon and rectum</td>
<td>0.91 (0.64–1.28) 0.57</td>
<td>1.08 (0.77–1.50) 0.52</td>
</tr>
</tbody>
</table>

Likelihood ratio test for interaction between primary tumor location and bevacizumab treatment in the two cohorts combinedb

<table>
<thead>
<tr>
<th>Hazard ratiob (95% CI) P</th>
<th>Hazard ratiob (95% CI) P</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.15 (0.03–1.15) 0.004</td>
<td></td>
</tr>
</tbody>
</table>

aPatients with multiple primary tumor locations (N = 15) were excluded from the analyses.
bAdjusted for age, sex, treating department, resected primary, prior adjuvant chemotherapy, number of metastatic sites, performance status, white blood cell count, and alkaline phosphatase level.

CAPEOXBEV, capecitabine, oxaliplatin, and bevacizumab; CAPEOX, capecitabine and oxaliplatin; CI, confidence interval.

There are important limitations to consider when interpreting our results. The data were collected retrospectively which could lead to bias and lower data quality. There was no central review done on CT scans which increases the uncertainty about progression timing. Yet, since OS, which was not affected by this uncertainty, showed the same prognostic division as PFS, this is unlikely to have had a significant effect on the results. Excluding cases with missing data did also not alter the results significantly.

Available, but there have been some reports comparing rectal and colon cancer. In the pivotal phase III study showing improved survival from adding bevacizumab to 5-fluorouracil and irinotecan (IFL), AVF2107 g, patients with rectal cancer experienced a markedly larger benefit from bevacizumab (HR 0.47) than patients with colon cancer (HR 0.74), with a median OS of 24.2 and 19.5 months, respectively, whereas the median OS was similar for the two locations in the placebo arm [5, 18]. In the Bevacizumab regimens: Investigation of Treatment Effects and Safety (BRiTE) observational study of patients treated with bevacizumab in combination with different chemotherapy regimens, a large difference in median OS between patients with rectal and colon cancer was found, median OS 29.2 versus 21.9 months [19]. A subgroup analysis of the AVEX study, a randomized study of capcitabine + bevacizumab versus capcitabine alone as first-line treatment of mCRC, which was recently presented, also showed a larger benefit of bevacizumab on PFS for patients with rectal (HR 0.41) and rectal + colon cancer (HR 0.22) than colon-only cancer (HR 0.67) [20]. In randomized phase III trials of combination chemotherapy without bevacizumab where primary tumor location was investigated for prognostic value, no difference in survival between patients with colon and rectal cancer was reported [4, 6, 21–24]. We observed a significantly longer OS for patients with rectal versus colon cancer when treated with chemotherapy and bevacizumab, but not when treated with chemotherapy alone, which is in accordance with the mentioned prospective trial data.
with primary tumors originating in the rectum and sigmoid colon. Other VEGF-targeting agents such as aflibercept and regorafenib also might exhibit location-dependent efficacy, and we urge investigators of targeted agents to stratify for tumor location in their analyses. Because of the retrospective and exploratory nature of the present study, this finding should be interpreted as hypothesis-generating only, and it needs to be validated in data from completed randomized trials.

funding
This work was supported by Roche and the Herlev Hospital Research Foundation.

disclosure

references