Cetuximab plus cisplatin–5-fluorouracil versus cisplatin–5-fluorouracil alone in first-line metastatic squamous cell carcinoma of the esophagus: a randomized phase II study of the Arbeitsgemeinschaft Internistische Onkologie


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Background: This study assessed the activity of the mAb cetuximab in combination with cisplatin and 5-fluorouracil (5-FU) in advanced esophageal squamous cell carcinoma.

Patients and methods: For a maximum of six 29-day cycles, patients received cisplatin 100 mg/m², day 1, plus 5-FU 1000 mg/m², days 1–5 (CF), either alone or in combination with cetuximab (CET–CF; 400 mg/m² initial dose followed by 250 mg/m² weekly thereafter). The primary end point was tumor response. Tumor material was obtained for analysis of KRAS mutation status.

Results: Sixty-two eligible patients were included, 32 receiving CET–CF and 30 CF. Cetuximab did not exacerbate grade 3/4 toxicity, except for rash (6% versus 0%) and diarrhea (16% versus 0%). The overall response rate according to RECIST criteria was 19% and 13% and the disease control rate 75% and 57% for the CET–CF and CF arms, respectively. With a median follow-up of 21.5 months, the median progression-free survival was 5.9 and 3.6 months and median overall survival 9.5 and 5.5 months for CET–CF and CF, respectively. No KRAS codon 12/13 tumor mutations were identified in 37 evaluated samples.

Conclusion: Cetuximab can be safely combined with CF chemotherapy and may increase the efficacy of standard CF chemotherapy.

Key words: cetuximab, chemotherapy, esophageal cancer, KRAS mutation, squamous cell carcinoma

Introduction

Esophageal cancer is characterized by a poor prognosis, with around 80% of patients presenting with advanced disease at the time of diagnosis. Systemic chemotherapy with cisplatin and 5-fluorouracil (5-FU) is the most commonly used first-line regimen for advanced esophageal squamous cell carcinoma (ESCC). However, response rates are low at 15%–45% and median survival is usually shorter than 8 months [1–3].

Therefore, novel therapies are urgently needed to improve outcomes in this setting. High levels of expression of the epidermal growth factor receptor (EGFR) have been detected in 50%–70% of ESCCs [4–6] and have been correlated with prognosis [5–7]. Activated EGFR signals via the RAS, ERK1/2, PI3K/Akt and STAT pathways and may result in chemotherapy resistance, angiogenesis and enhanced tumor proliferation [8, 9]. In addition, preclinical data showed that cetuximab enhanced the antitumor effect of chemotherapy and, in particular, augmented the activity of cisplatin [10, 11]. Therefore, it was reasoned that EGFR blockade may be an effective therapeutic strategy.
Cetuximab, an immunoglobulin G1 mAb that specifically targets the EGFR, has shown efficacy in the treatment of colorectal cancer [12], non-small-cell lung cancer (NSCLC) [13] and both recurrent/metastatic [14–17] and locally advanced head and neck squamous cell carcinoma (HNSCC) [18]. Due to the proven antitumor activity of cetuximab in HNSCC which has high levels of EGFR expression and comparable tumor biology compared with ESCC, we hypothesized that this agent may be active in the treatment of advanced ESCC.

The current phase II study therefore investigated the activity and safety of cetuximab added to standard cisplatin–5-FU therapy (CET–CF) in patients with advanced ESCC previously not treated for advanced disease. Those patients with overt disease progression under cisplatin–5-FU (CF) were to be allowed to receive cetuximab, with or without cisplatin–5-FU.

**patients and methods**

This multicenter, open-label, noncomparative randomized phase II study was conducted at 12 institutions in Germany. It was coordinated within the study network of Arbeitsgemeinschaft Internistische Onkologie. The protocol was approved by the ethics committee for human research at the Technische Universität München, Munich, Germany, and conformed to the principles of the Declaration of Helsinki and its subsequent amendments.

**eligibility**

Patients aged ≥18 years, with histologically confirmed, EGFR-expressing, nonresectable, advanced ESCC, who had not received (neo)adjuvant chemotherapy within 6 months of study entry or prior chemotherapy for recurrent or metastatic disease were eligible. They required an Eastern Cooperative Oncology Group performance status (ECOG PS) of one or less, creatinine clearance >70 ml/min, an adequate hepatic and bone marrow function and a unidimensionally measurable lesion ≥1 cm in diameter detected by computed tomography (CT) scan. A second malignancy, uncontrolled infection, neuropathy grade ≥1 and pregnancy or lactation excluded participation. All patients gave written informed consent.

**EGFR expression analysis**

Before randomization, EGFR expression was evaluated locally in a tumor specimen using a standardized immunohistochemistry assay (EGFR pharmDx; Dako, Glostrup, Denmark). Positive staining was defined as any membrane staining above background level (defined as the level noted in a negative control sample) in ≥21% cancer cells of any intensity; with 1+ equating to faint or barely perceptible membrane staining, 2+ indicating weak to moderate staining of the complete cell membrane and 3+ indicating strong staining of the complete cell membrane.

**KRAS mutational analysis**

Screening of tumor DNA for mutations in codon 12 or 13 of KRAS was carried out centrally. Before DNA extraction, tumor tissue from previous diagnostic or surgical procedures (frozen or paraffin embedded) was micro dissected to assure at least 60% tumor cell content of the analyzed sample. PCR amplification, sequencing and mutation detection were carried out according to Brink et al. [19].

**treatment**

Patients were randomized centrally 1 : 1, without stratification, to receive a 29-day cycle: cisplatin 100 mg/m² over 60 min on day 1, with standard antiemetic prophylaxis and pre- and postcisplatin hydration, followed by 5-FU 1000 mg/m²/day as a continuous 24 h i.v. infusion from days 1–5, either alone (CF) or with cetuximab, administered as an initial dose of 400 mg/m² on day 1 over 120 min, followed by weekly doses of 250 mg/m² over 60 min (CET–CF). Patients received chemotherapy until disease progression or the occurrence of unacceptable toxicity, for up to six cycles. Patients who showed disease progression in the CF arm were permitted to receive cetuximab plus chemotherapy or cetuximab alone, provided that they continued to meet eligibility criteria.

**dose modifications**

Toxicity was graded according to National Cancer Institute—Common Toxicity Criteria version 3.0. Dose modifications following the occurrence of cetuximab- and chemotherapy-related toxicity were prespecified in the protocol.

**study assessments**

Before randomization, a complete medical history was taken, tumor-related symptoms were recorded and a full body examination was carried out. Complete blood cell count (CBC), blood chemistry analyses, electrocardiography and tumor assessment were carried out. CBC was evaluated weekly and electrolyte levels every second week. Patients were assessed every cycle for potential adverse events (AEs) and disease-related signs and symptoms. Tumor assessments by CT scans of the chest and abdomen were carried out at baseline and every 8 weeks according to RECIST guidelines [19]. Responses were to be confirmed by repeated assessments carried out no less than 4 weeks apart. After six chemotherapy cycles, patients were monitored every 12 weeks until disease progression.

**statistical analysis**

Patients were considered assessable for response and toxicity if they had received at least one dose of treatment. The primary end point of the study was the confirmed objective response rate (ORR). The study was designed as a Gehan two-stage trial and assumed a response rate of ≥40% in the CET–CF arm. With a power of 90%, this resulted in a sample size of five patients for the first stage. The size of the second stage was determined by the observed number of responses and by the prespecified precision of 10%. Since one response was observed in the first stage, the study could be continued to a total of 25 patients in the CET–CF arm. Secondary end points included overall survival (OS), progression-free survival (PFS), duration of response, time to treatment failure (TTF) and safety. Analysis of PFS and OS was carried out using the Kaplan–Meier product-limit method. Comparisons between groups of patients were made by log-rank tests. Median survival and hazard ratios (HRs) calculated by Cox proportional hazards model are reported, with 95% confidence intervals (CIs). PFS was determined from the day of study assignment to the date of progression, death or last contact. TTF was determined from the day of first chemotherapy infusion until discontinuation of treatment for any reason. Patients who had not progressed at the time of the final analysis were censored at the date of their last tumor assessment. OS was calculated from the day of assignment to death. Patients alive at the final survival analysis were censored using the last contact date.

Statistical analysis was carried out using SPSS software (version 15.0; SPSS, Inc., Chicago, IL). Ninety-five percent CIs were calculated using StatXact (version 5; Cytel, Inc., Cambridge, MA). All statistical analyses were carried out at a 5% level of significance.

**results**

**patient characteristics**

A total of 66 patients were registered from 14 December 2004 to 13 December 2006. Two patients died and two patients had
a deterioration of their PS before therapy start and were therefore excluded from the outcome analyses, leaving 62 eligible for inclusion. Of the 62 eligible cases, 32 patients were randomly assigned to the CET–CF arm and 30 to the CF arm (Figure 1). At the time of final analysis, the median follow-up time was 21.5 months. Five patients in the CF arm who experienced disease progression crossed over to receive CET–CF or cetuximab alone.

Table 1 summarizes the patient and disease characteristics at baseline. Both groups were well balanced with respect to age, ECOG PS, tumor differentiation and intensity of EGFR staining. However, there was a marked difference in the gender balance between the arms. More than 30% of patients in each arm had either prior surgery or radiotherapy or both. In contrast, only 13% of patients in each arm had received prior systemic chemotherapy.

**KRAS mutation analysis**

Tumor samples from 37 of 62 patients were analyzed for KRAS mutation status: the remaining 25 samples were either missing (n = 13) or considered to contain insufficient tumor cell content (n = 12). Among the samples tested, no activating point mutations of the KRAS gene were detected.

**treatment administration**

In 62 patients, a median of four cycles (range one to eight cycles) per patient and three cycles (range one to nine cycles) per patient were administered in the CET–CF and the CF arms, respectively. The median cumulative dose per patient was 811 versus 659 mg for cisplatin and 41 440 mg versus 29 500 mg for 5-FU in the CET–CF versus CF arm, respectively. The median cumulative dose per patient for cetuximab was 8690 mg. Early treatment discontinuation was necessary in 22 patients (69%) in the CET–CF arm and in 25 patients (83%) in the CF arm. The most common reason for treatment discontinuation was tumor progression (CET–CF, 41%; CF, 64%). Other reasons for treatment discontinuation in the CET–CF versus CF arm were toxicity 23% versus 20%, mainly renal; patient preference 27% versus 12% and unknown 9% versus 4%, respectively. One patient on CET–CF had a grade 4 infusion-related reaction and was not retreated with cetuximab. At least one dose attenuation to <80% of the initial drug dose was required in 10 of 32 (31%) and 10 of 30 (33%) patients in the CET–CF and the CF arms, due to grade 3/4 toxic effects, including reductions for cisplatin in 8 of 32 patients (25%) and 9 of 30 (30%) patients and for 5-FU in 8 of 32 patients (25%) and 7 of 30 (23%) patients, respectively. Dose reductions of either cisplatin or 5-FU were due to grade 3/4 neutropenia in two (6%) and three (10%) patients, grade 3/4 diarrhea in two (6%) and zero patients, renal impairment in three (9%) and five patients (17%) and mucositis in three (9%) and two patients (7%) for the CET–CF and CF arms, respectively. There were two deaths in the CET–CF arm and nine deaths in the CF arm during treatment or within 30 days after the final dose. All, except for one death due to septicemia in the CF arm, were attributed to disease progression.

Second- or third-line chemotherapy was given in 12 of 32 patients (38%) and 6 of 30 patients (20%) in the CET–CF and CF arms, respectively.

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**Figure 1.** Consolidated Standards of Reporting Trials diagram. *Because of death (one patient in each arm) and rapid disease progression with deterioration of general health status (two patients in CF arm) before treatment was initiated. CET–CF, cetuximab, cisplatin and fluorouracil; CF, cisplatin and fluorouracil.
Table 1. Patient and tumor characteristics at baseline

<table>
<thead>
<tr>
<th>No. of patients</th>
<th>CET–CF arm</th>
<th>CF alone arm</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gender</td>
<td>Male/female</td>
<td>Male/female</td>
</tr>
<tr>
<td>No. of patients</td>
<td>32</td>
<td>30</td>
</tr>
<tr>
<td>Median age (range), years</td>
<td>61 (40–76)</td>
<td>62 (40–74)</td>
</tr>
</tbody>
</table>

Table 2. Hematological and nonhematological toxic effects (National Cancer Institute—Common Toxicity Criteria, Version 3.0)

<table>
<thead>
<tr>
<th>Adverse event</th>
<th>CET–CF arm (n = 32)</th>
<th>CF alone arm (n = 30)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Any grade</td>
<td>Grade 3/4</td>
</tr>
<tr>
<td>Hemoglobin</td>
<td>n</td>
<td>%</td>
</tr>
<tr>
<td>Neutrophils</td>
<td>27</td>
<td>84</td>
</tr>
<tr>
<td>Platelets</td>
<td>21</td>
<td>66</td>
</tr>
<tr>
<td>Diarrhea</td>
<td>13</td>
<td>41</td>
</tr>
<tr>
<td>Nausea</td>
<td>22</td>
<td>69</td>
</tr>
<tr>
<td>Emesis</td>
<td>15</td>
<td>47</td>
</tr>
<tr>
<td>Fatigue</td>
<td>20</td>
<td>63</td>
</tr>
<tr>
<td>Fever</td>
<td>7</td>
<td>22</td>
</tr>
<tr>
<td>Sensory neuropathy</td>
<td>6</td>
<td>19</td>
</tr>
<tr>
<td>Hand–foot syndrome</td>
<td>7</td>
<td>22</td>
</tr>
<tr>
<td>Acne-like rash</td>
<td>22</td>
<td>69</td>
</tr>
<tr>
<td>Infusion-related reaction</td>
<td>3</td>
<td>9</td>
</tr>
</tbody>
</table>

4Infusion-related reaction is a composite category including the Medical Dictionary for Regulatory Activities 8.0 terms: infusion-related reaction, hypersensitivity, anaphylactic reaction, anaphylactic shock, anaphylactoid reaction, dyspnea*, pyrexia* and chills* (*only taken into account when occurring on the first day of the first cetuximab administration). These composite category data are included to provide additional safety information of relevance to cetuximab administration. CET–CF, cetuximab, cisplatin and fluorouracil; CF, cisplatin and fluorouracil.

Table 3. Antitumor activity (n = 62)

<table>
<thead>
<tr>
<th>Response</th>
<th>No. of patients (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>CET–CF (n = 32)</td>
</tr>
</tbody>
</table>

Best overall response
- CR | 0 (0) | 1 (3) |
- PR | 11 (34) | 8 (27) |
- SD | 13 (41) | 8 (27) |
- Progressive disease | 6 (19) | 10 (33) |
- Not evaluable | 2 (6) | 3 (10) |
- Objective response rate, CR + PR [95% CI] | 11 (34) [19–53] | 9 (30) [15–49] |
- Disease control rate, CR + PR + SD [95% CI] | 24 (75) [57–87] | 17 (57) [37–75] |
- Confirmed response rate (RECIST) [95% CI] | 6 (19) [7–36] | 4 (13) [4–31] |

CET–CF, cetuximab, cisplatin and fluorouracil; CF, cisplatin and fluorouracil; CR, complete response (disappearance of all lesions); PR, partial response; SD, stable disease; CI, confidence interval.

Reasons were either missing or poor quality scans in one patient in the CF arm and early treatment discontinuation (<4 weeks of treatment) in one and two patients in the CET–CF and CF arm, respectively. The ORR in the ITT population for patients receiving CET–CF was 34% (95% CI 19% to 53%) and 30% (95% CI 15% to 49%), while for patients in the CF arm (P = 0.79). The confirmed ORR was 19% (95% CI 7% to 36%) in the CET–CF arm and 13% (95% CI 4% to 31%) for patients in the CF only arm (P = 0.73). The disease control rate in the CET–CF arm and the CF arm were 67% (95% CI 49% to 81%) and 39% (95% CI 22% to 57%), respectively.
CET–CF arm was 75% and 57% in the CF arm (\(P = 0.18\)). Median time to response was 8 and 8 weeks (range 7–11 and 4–16) and median duration of response was 15 and 18 weeks (range 7–56 and 8–27; \(P = 0.76\)) for the CET–CF and the CF arms, respectively. The response rate for the 16 patients in the CET–CF arm who developed grade 2 or 3 skin toxicity was 44% (seven responders) compared with 31% (five responders) among the 16 patients who had grade 1 or no skin toxicity. This difference was not statistically significant (\(P = 0.74\)).

PFS and survival
At a median follow-up of 21.5 months, 46 (74%) patients had died, 24 (52%) in the CET–CF and 22 (48%) in the CF arm. PFS tended to be longer in the CET–CF arm (Figure 2A) without reaching statistical significance (\(P = 0.21\)). Median PFS was 5.9 months (95% CI 3.8–8.0 months) for patients in the CET–CF arm and 3.6 months (95% CI 1.0–6.2 months) for patients treated with CF alone. In addition, a significant difference was not observed in TTF [HR 0.71 (95% CI 0.39–1.28); \(P = 0.25\)]. Median TTF was 3.4 months (95% CI 2.6–4.3) in the CET–CF arm and 1.6 months (95% CI 0–3.6) in the CF arm.

Survival also tended to be longer in the CET–CF arm without reaching statistical significance (Figure 2B). Median OS was 9.5 months (95% CI 8.4–10.6 months) for the CET–CF arm and 5.5 months (95% CI 1.9–9.1 months) for the CF arm (\(P = 0.32\)). The estimated 2-year survival rate was 28% for patients treated with CET–CF compared with 18% for patients treated with chemotherapy alone. Of note, no significant difference was seen in median PFS, OS and ORR between patients who had received prior treatment (surgery and/or radiotherapy and/or chemotherapy) and those patients who were treatment naive. Furthermore, Cox hazards regression modeling of PFS and OS on treatment group, controlling for gender, did not change the results compared with the analysis by treatment group alone.

crossover population
From May 2005 until June 2006, five patients whose tumor had progressed on CF, two women and three men, crossed over to receive CET–CF or cetuximab alone. No dose reductions and delays became necessary and side-effects were manageable. The median time to crossover was 3.7 months (range 2.9–4.1). A median of 12 weeks (range 8–24) of additional therapy was delivered to the five crossover patients.

The efficacy analysis showed a disease control rate [partial response (PR; 30% decrease of the sum of the longest diameter (one dimension)) or stable disease (SD; Neither PR nor PD criteria met)] of 60%. Two patients received cetuximab monotherapy with one achieving a PR and the other an SD. Three patients received CET–CF. Two patients showed progressive disease after two cycles of therapy and one patient achieved a PR. For the five crossover patients, median PFS from time of crossover was 6.8 months (95% CI 5.9–7.6) and median OS was 7.1 months (95% CI 5.4–8.8).

discussion
Given that cisplatin combined with 5-FU is the treatment of choice for patients with ESCC, we sought to increase efficacy by adding cetuximab to this regimen. The unconfirmed ORR of 34% in the CET–CF arm is in line with previously reported results for cisplatin plus 5-FU [1]. The confirmed response rate, however, was only 19%. Therefore, disappointingly, the CET–CF arm did not meet the primary objective of demonstrating a \(\geq 40\%\) response rate. However, evaluation of response rates alone may underestimate the overall treatment
effect of targeted therapies. Previous studies have shown that blockade of EGFR may induce disease stabilization rather than tumor shrinkage [20]. Our findings were in agreement with this notion, with a high number of patients (75%) experiencing disease control in the CET–CF arm compared with little more than 50% with CF alone. However, several newer chemotherapy agents have shown impressive response rates of 40%–66% and survival rates ranging between 8 and 15 months [21–24] in phase II trials. Nevertheless, it should be noted that many of these studies were nonrandomized and included heterogeneous patient populations. Moreover, the consistent use of confirmed responses according to RECIST guidelines remains unclear in the majority of these studies. Because outcomes can be markedly influenced by patient selection in advanced ESCC, we decided to use a randomized and controlled trial design to minimize bias related to patient selection [25]. Due to the limited sample size, this study does not allow for robust statistical comparisons between the arms in relation to PFS and OS.

The median PFS (5.9 versus 3.6 months) and the median OS (9.5 versus 5.5 months) both favored the CET–CF combination, although it must be noted that the increased OS could be due to the higher rate of second-line therapies administered in the CET–CF arm. An imbalance of gender distribution between the two study arms could be another potential source of bias. Thus, no significant impact of gender with regard to survival and PFS, respectively, was observed. However, the poor survival (PFS 3.6 months and OS 5.5 months) as well as the low confirmed response rate (13%) in the control group reflect the enrollment of a patient population with a particularly poor prognosis. In view of this factor, the efficacy data for the CET–CF arm are encouraging.

The rates and types of AEs in our patients were consistent with those expected from the individual agent. Moreover, there was no evidence that cetuximab aggravated the known toxic effects of CF. However, a significantly higher incidence of grade 3/4 AEs was noted in the CET–CF compared with the CF arm, possibly related to the higher number of treatment cycles administered in the CET–CF arm compared with the CF arm, an assertion supported by the similarity in rates of early treatment discontinuations due to toxicity and dose reductions. Nevertheless, more than twice as many patients in the CET–CF arm compared with the CF alone arm discontinued treatment on request, mainly due to cetuximab-related skin reactions, while in the CF, significantly more patients discontinued treatment due to disease progression. Notably, one patient had a serious infusion-related reaction following cetuximab administration, leading to discontinuation of treatment. However, no deaths were reported to be related to cetuximab.

In addition to clinical parameters, we investigated the mutational status of the EGFR-signaling effector KRAS. Similar to NSCLC [26] and colorectal cancer [27, 28], KRAS mutations in esophageal cancer may negatively correlate with the response to anti-EGFR therapy [27]. In line with previous observations, no tumor KRAS mutations were detected in the current study [27].

In conclusion, within the limits of the trial design and the small patient numbers eligible for such a trial, the findings from this study indicate that the triple combination regimen (CET–CF) may provide a useful therapeutic approach and merits additional investigation. The high rate of treatment discontinuation due to renal toxicity in both treatment arms may necessitate a reduced cisplatin dose in clinical practice. The greatest benefit may ultimately arise from integration of cetuximab into multimodal treatment approaches.

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Authors’ disclosure of potential conflicts of interest:
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