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ORIGINAL ARTICLE

A proof of concept trial of the anti-EGFR antibody mixture Sym004 in patients with squamous cell carcinoma of the head and neck

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Abstract
Purpose The purpose of the trial was to assess the efficacy and tolerability of Sym004, a novel 1:1 mixture of two chimeric monoclonal antibodies (992 and 1024) targeting non-overlapping epitopes of the anti-epidermal growth factor receptor (EGFR), in patients with squamous cell carcinoma of the head and neck (SCCHN).
Methods Incurable, recurrent and/or metastatic SCCHN patients with acquired resistance to anti-EGFR monoclonal antibody-containing treatment received weekly infusions of 12 mg/kg Sym004 until disease progression or unacceptable toxicity.
Results Among the 26 patients treated with Sym004, the proportion of patients alive without disease progression at 6 months was 12 % (95 % CI 1–39 %). The median duration of progression-free survival was 82 days (95 % CI 41–140 days). Of 19 patients evaluable for response, eight showed a decrease in the sum of the largest diameter in their target lesions (median 11 %; range 7–27 %). The best overall response was stable disease in 13 patients (50 %). Paired biopsies showed a significant down-regulation of EGFR in both skin and tumors following exposure to Sym004. All patients had EGFR-related adverse events, including grade 3 skin toxicities and grade ≥3 hypomagnesemia reported in 13 (50 %) and 10 (38 %) of 26 patients, respectively. One event fulfilling the protocol-defined criteria for infusion-related reactions (grade 2) was reported. No anti-drug antibodies were detected.
Conclusions The marked EGFR down-regulation shown in target tissues supports the proposed mechanism of action of Sym004. This trial revealed modest anti-tumor activity of Sym004 in extensively pretreated advanced SCCHN patients.

Keywords Head–neck cancer · EGFR resistance · Anti-EGFR antibody · Treatment · Phase 2a · Sym004

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Introduction

Squamous cell carcinoma of the head and neck (SCCHN) still poses a serious condition to treat, especially once recurrent or metastatic. Despite the multimodal treatment, local recurrence or/and distant metastases will occur in almost half of patients with locally advanced disease. The first-line palliative treatment consists of platinum-based chemotherapy in combination with cetuximab, a monoclonal antibody (mAb) targeting the epidermal growth factor receptor (EGFR). Overall, patients' prognoses remain poor despite a median overall survival (OS) of up to 11 months following anti-EGFR mAb treatment in combination with platinum-based chemotherapy, as compared to as low as 7 months following chemotherapy [1, 2]. For patients not eligible to chemotherapy or with disease progression after platinum-based therapy, no treatment option is available with a demonstrated improvement in survival [3–5].

Only a fraction (6–13 %) of patients treated with anti-EGFR mAb monotherapy show objective tumor responses, and disease stabilization in this setting is seen in only 42–56 % [5, 6]. Irrespective of their response, all patients will ultimately acquire resistance to available anti-EGFR treatment. As a possible mechanism of resistance to anti-EGFR treatment, activation of tyrosine kinase receptors, including human epidermal receptor (HER)-2, HER-3 and MET, has been reported [7–9]. High levels of EGFR ligands such as amphiregulin have also been shown to promote resistance to EGFR-targeting agents [8].

Sym004 is a novel 1:1 mixture of two chimeric mAbs (992 and 1024) targeting non-overlapping epitopes in domain III of the EGFR [10, 11]. In our preclinical work, Sym004 has been shown to induce internalization and degradation of EGFR more efficiently than individual and reference mAbs by a mechanism involving EGFR crosslinking [10, 11]. Sym004-induced EGFR removal translated into superior inhibition of downstream signaling pathways and improved anti-tumor activity [10, 11].

The dose and schedule of Sym004 were selected based on the limited experience in the phase 1 dose escalation trial in patients with solid tumors (Trial registration ID: NCT01117428). In this trial, Sym004 was administered at doses from 0.4 to 12 mg/kg/week with a safety and toxicity profile similar to that expected from known EGFR antibodies [12, 13]. Although the formal maximum-tolerated dose was not reached in the trial, 12 mg/kg/week was the highest administered dose. The decision not to administer doses higher than 12 mg/kg/week was based on the short-term as well as the cumulative toxicity of Sym004. Furthermore, the selection of the 12 mg/kg/week dose for the present phase 2a trial was supported by the pharmacodynamic data in the phase 1 trial, which showed a complete removal of the EGFR at this dose. The present phase 2a trial (Trial registration ID: NCT01417936) aimed to assess the efficacy [as measured by progression-free survival (PFS)] and toxicity of Sym004 monotherapy in recurrent SCCHN patients previously treated with an anti-EGFR mAb.

Patients and methods

Trial design

This trial was an open-label, single arm, multicenter proof of concept trial. Eligible patients received intravenous Sym004 monotherapy (12 mg/kg every week) until disease progression, unacceptable toxicity or the wish of the patient to withdraw from the trial.

Patients

Eligible patients had histologically confirmed SCCHN (no patients with nasopharyngeal carcinoma or known brain metastases were enrolled), Eastern Cooperative Oncology Group (ECOG) performance status 0–1, recurrent and/or metastatic disease not amenable to curative treatment, measurable disease (according to Response Evaluation Criteria in Solid Tumors (RECIST) [14]) with tumor lesions accessible for repeated biopsies, previous anti-EGFR mAb-containing palliative treatment and progressive disease according to RECIST 1.1 criteria at inclusion. Patients had to have (1) a documented clinical benefit or response for at least 8 weeks (partial response, complete response or stable disease) to anti-EGFR mAb in combination with either chemotherapy or radiotherapy or as monotherapy and (2) a documented progressive disease during or within 12 weeks following anti-EGFR mAb treatment. Patients were allowed to have had up to two lines of previous chemotherapy in the palliative setting. Patients needed to have adequate hematologic, renal and hepatic organ function.

The clinical and translational parts of the trial were approved by the Independent Ethics Committees and the Health Authorities and were conducted in accordance with the Declaration of Helsinki (Seoul 2008). Written informed consent was obtained from each patient. It was prospectively planned to perform translational research, and patients gave their informed consent for repeated biopsies and blood collection.

Treatments

Sym004 was administered as an intravenous infusion at a maximum rate of 10 mg/min once every week. A premedication schedule included a glucocorticoid (before

the first two infusions) as well as an anti-histamine and an anti-pyretic (before the first four infusions). Prophylaxis of rash was allowed at the investigator's discretion with minocycline, doxycycline or similar, preferably prior to first infusion. Other skin toxicity prophylaxis regimens (e.g., topical creams) could be applied, as per local practice.

Endpoints and evaluation of outcomes

The primary endpoint was the rate of PFS (by central evaluation) at 6 months (24 weeks). Secondary endpoints were the frequency and nature of adverse events, best overall response, time to progression (TTP), OS, pharmacokinetics and changes in skin and tumor biomarker expression from baseline.

Adverse events were graded according to Common Toxicity Criteria for Adverse Events (CTCAE) version 4.02. Tumors were evaluated by computerized tomography (CT) or magnetic resonance imaging using RECIST version 1.1 against a baseline assessment performed within 4 weeks before the first dose. Re-assessments were performed after the first 6 and 12 weeks of treatment and thereafter every 8 weeks. Relative to the first infusion of Sym004, TTP, PFS and OS were calculated as the duration until event (progression, progression or death and death, respectively). The average relative dose intensity of Sym004 was calculated as the actual dose administered in the actual treatment period divided by the dose theoretically planned for that treatment period, multiplied by 100.

For pharmacokinetic and immunogenicity analyses, serial blood samples were collected to measure Sym004 and anti-Sym004 antibodies, respectively, using enzyme-linked immunosorbent assays.

Tissue samples

Pharmacodynamic effects of Sym004 were explored in biopsies from skin ($N = 18$) and tumor ($N = 11$) lesions, obtained by needle punch biopsies or excisional biopsies at baseline and 4 weeks after the first Sym004 infusion, i.e., prior to the planned fifth infusion. Semiquantitative immunohistochemistry studies for EGFR expression were performed at HistoGeneX, Belgium. Formalin-fixed paraffin-embedded samples were de-paraffinated and treated with protease for 8 min before incubation with anti-EGFR (clone 3C6, Ventana) for 32 min. Following hematoxylin staining, EGFR staining was visualized with ultraView™ Universal DAB (3,3-Diaminobenzidine) Detection Kit (Ventana) and a bluing reagent (Ventana). Staining intensity was scored as 0, 1, 2 or 3 corresponding to the presence of negative, weak, intermediate and strong brown staining, respectively. An EGFR histoscore (H-score) was calculated as 3 × percentage of cells with strong staining + 2 × percentage of cells with intermediate staining + percentage of cells with weak staining, giving a H-score range from 0 to 300.

Available formalin-fixed paraffin-embedded tumor samples were analyzed at HistoGeneX, Belgium, by polymerase chain reaction for the presence of (1) 18 different human papilloma virus (HPV) subtypes [15] and (2) *EGFRvIII* messenger ribonucleic acid (Roche Diagnostics). *MET* gene copy number in individual tumor cells was determined by fluorescent in situ hybridization (Kreatech Diagnostics).

Statistical methods

The full analysis set (FAS) comprised all patients who had been exposed to at least one dose of trial drug. The FAS was used for the evaluation of all efficacy and safety endpoints. For the assessment of pharmacokinetic and pharmacodynamic endpoints, only patients with available samples were considered.

To describe the 6-month PFS rate (primary endpoint), the assumptions for the sample size calculation were based on a PFS rate of 7.3 % at 6 months (null hypothesis) in findings from a phase 3 trial performed in a similar population [5]. Assuming a 6-month PFS rate of 25 %, a sample size of 25 patients would give a power of 0.79 on a 5 % significance level to detect an increased PFS rate. PFS time was estimated using the Kaplan–Meier method and presented with corresponding two-sided 95 % confidence intervals (CIs). Due to the nature of disease progression, the primary endpoint was supplemented by descriptive statistics of median PFS including two-sided 95 % CIs. Descriptive standard statistics were used to analyze secondary endpoints.

The following pharmacokinetic endpoints were calculated for Sym004 following the first and fourth infusions: area under concentration–time curve from start of first infusion to 168 h ($AUC_{0–168h}$), area under concentration–time curve from start of first infusion to infinity ($AUC_{0–inf}$), half-life ($T_{½}$) and clearance (CL). Maximum concentration (C_{max}) and minimum concentration (C_{min}) were calculated using all data. C_{max} and C_{min} were derived from raw data, while $AUC_{0–168h}$, $AUC_{0–inf}$, CL and $T_{½}$ were estimated using non-compartmental modeling. For pharmacodynamic endpoints, a two-tailed Wilcoxon signed rank test for matched pairs was used to compare immunohistochemistry assessments from paired biopsies.

All statistical analyses were conducted using SAS 9.3 program, and differences between samples were considered significant when a p value below 0.05 was assessed.

Results

Patient characteristics

Patient allocation and disposition to treatment are shown in Fig. 1. Baseline characteristics of the FAS ($N = 26$) are described in Table 1, showing a SCCHN population with extensive pretreatment. All patients had been exposed to anti-EGFR mAb treatment (two patients had been exposed twice to anti-EGFR mAb treatment in different lines of treatment, and one patient had received an investigational anti-EGFR tyrosine kinase inhibitor in addition to prior anti-EGFR mAb treatment). In 24 patients (92 %), disease progression had been assessed within 21 days of the last dose of anti-EGFR mAb treatment. The median time between progression on previous anti-EGFR treatment and first dose of Sym004 was 42 days (interquartile range 23–118 days). Only one patient had HPV-positive tumor, and no *EGFRvIII* mRNA was detected in tumor specimens (Table 1). No *MET* amplifications were found.

Anti-tumor activity

The proportion of patients alive and free of progression at 6 months was 12 % (95 % CI 1–39 %) in the FAS. Median PFS and TTP were 82 days (95 % CI 41–140 days) and 85 days (95 % CI 42–147 days), respectively. Of 19 patients evaluable for response, eight had a decrease in the sum of the largest diameter of their target lesions from baseline (median 11 %; range 7–27 %)

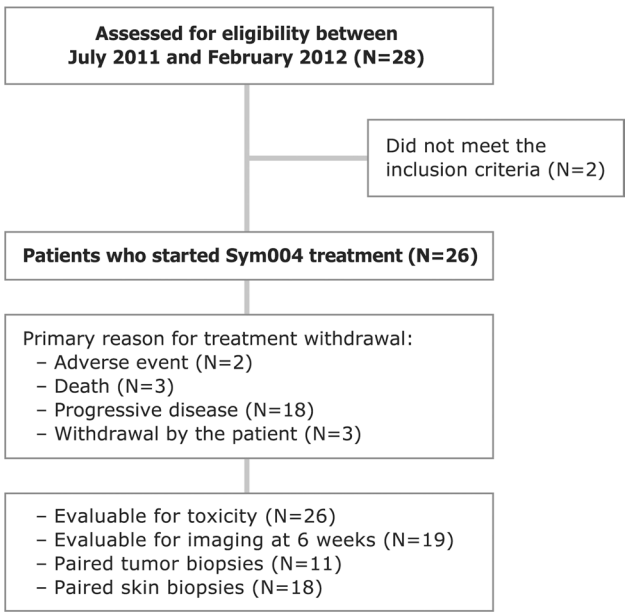

Fig. 1 CONSORT diagram of trial Sym004-02

Table 1 Patient characteristics

Characteristic	*N* (%)
Full analysis set (FAS)	26 (100)
Age (*years*)	
Median	60
Range	42–87
Sex	
Female	3 (12)
Male	23 (88)
ECOG	
0	3 (12)
1	23 (88)
Alcohol history	
Yes	18 (70)
No	4 (15)
Unknown	4 (15)
Tobacco history	
Yes	25 (96)
Unknown	1 (4)
Primary tumor location	
Larynx	7 (26)
Hypopharynx	3 (12)
Oropharynx	13 (50)
Others	3 (12)
Presence of distant metastases at inclusion	13 (50)
EGFR variant III	
Absent	21 (81)
Present	0
Not available	5 (19)
Human papillomavirus	
HPV positive	1 (4)
HPV negative	20 (77)
Not available	5 (19)
Prior curative therapy as a part of the multimodality treatment	
Radiation therapy	11 (42)
Chemoradiation	11 (42)
Surgery	13 (50)
No curative treatment	4 (15)
Number of previous treatment lines for palliation	
Treated with only one line of palliative treatment	7 (27)
Treated with two or more lines of palliative treatment	19 (73)
Progression on prior anti-EGFR mAb treatment	
Within 21 days of last dose	24 (92)
Type of anti-EGFR mAb received for palliation	
Cetuximab	25 (96)
Zalutumumab	1 (4)

N (%) = number of patients with events (percent of patients)

ECOG Eastern Cooperative Oncology Group, *EGFR* epidermal growth factor receptor, *HPV* human papillomavirus, *mAb* monoclonal antibody

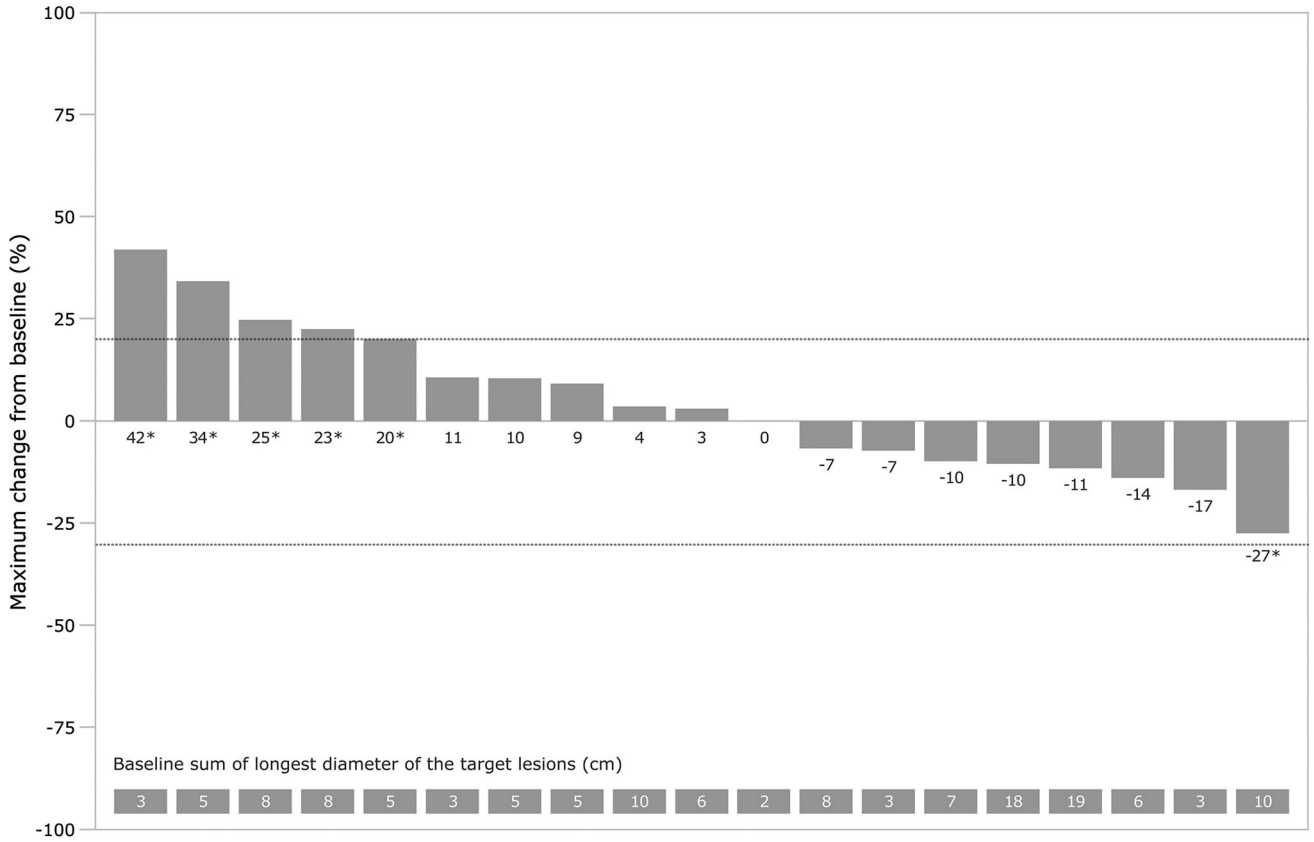

Fig. 2 Maximum relative change in the sum of the longest diameters of target lesions from baseline in patients evaluable for response by central review. The *bars* indicate the largest percentage change in target lesions from baseline, and the corresponding individual percentages of change are shown below the *bars*. The *lower dashed horizontal line* indicates a 30 % reduction from baseline; *a* > 30 % reduction from baseline is defined as partial response as per RECIST 1.1. Progressive disease, defined as *a* > 20 % increase from baseline, is indicated by the *upper horizontal line*; *asterisk* indicates patients with progressive disease as overall best response

(Fig. 2). The best overall response was stable disease in 13 patients (50 %). For seven patients, data on response were missing due to withdrawal of consent ($N = 3$), death prior to post-baseline imaging ($N = 2$) or non-evaluable disease status ($N = 2$). Median OS was 156 days (95 % CI 86–202 days), and the 24-week survival estimate was 42 % (95 % CI 22–60 %).

Exposure and adverse events

The median treatment duration was 7.6 weeks (range 2–30 weeks) with a mean relative dose intensity of 78 % (standard deviation 19 %) (Table 2). Nineteen patients received 12 mg/kg throughout the trial period. Sym004 was reduced to 9 mg/kg in three patients and to 6 mg/kg in four patients due to skin toxicities. Two patients stopped treatment due to adverse events: one for fatigue grade 3 and one for a device-related infection (port infection) grade 3 (Table 2). No anti-drug antibodies were detected.

A summary of adverse events is shown in Table 2. Adverse events assessed as related to Sym004 and occurring in more than 10 % of the patients are shown in Supplementary Table 1, together with all related serious adverse events. The most frequent adverse events were skin toxicity events (92 %), hypomagnesemia (65 %), fatigue (39 %) and diarrhea (35 %). Grade 3 skin toxicities were recorded in 13 patients (50 %); no grade 4 skin toxicities were recorded (Table 2).

No patients discontinued Sym004 due to hypomagnesemia, which was generally managed by oral or intravenous substitution. No cardiac events related to hypomagnesemia were reported. One grade 2 infusion-related reaction was confirmed and met the prespecified criteria of occurrence within 2 h and investigator's assessment as Sym004 related. According to the investigator, one potentially Sym004-related grade 5 adverse event was recorded (fatal pharyngeal tumor bleeding extra muros secondary to tumor erosion).

Table 2 Summary of adverse events and exposure, full analysis set

Variable	*N* (%)
Adverse events	
Any adverse event[a]	26 (100)
Any grade ≥3 adverse event	24 (92)
Any serious adverse event	20 (77)
Drug-related fatal adverse events	1 (4)
Sym004 discontinuation due to adverse event[b]	2 (8)
Skin toxicity adverse events[c]	24 (92)
Grade 3	13 (50)
Hypomagnesemia	17 (65)
Grade ≥3	10 (38)
Fatigue	10 (39)
Grade ≥3	5 (19)
Diarrhea	9 (35)
Grade ≥3	0
Sym004 exposure	
Treatment duration (weeks)	
Median	7.6
Range	2–30
Relative dose intensity (%)	
Mean (SD)	78 (19)
Dose reduced, *N* (%)	7 (27)
Treatment interrupted, *N* (%)	19 (73)

N (%) = number of patients with events (percent of patients)

[a] Adverse events were recorded from first dose until the first follow-up visit

[b] Excluding four cases of disease-related death and disease progression reported as adverse events

[c] No grade 4 event was observed. Includes adverse events with the following preferred terms: dermatitis acneiform, dry skin, erythema, face edema, folliculitis, nail infection, paronychia, pruritus, rash, rash erythematous, rash maculo-papular, xerosis

Pharmacokinetics and pharmacodynamics

After the fourth infusion of 12 mg/kg Sym004, the geometric mean AUC increased by approximately 1.3-fold as compared to first infusion. The serum $T_{1/2}$ of Sym004, estimated by the compilation of serum elimination curves, was 3 days after the first infusion and 6 days after the fourth infusion, respectively (Supplementary Table 2). In patients who received uninterrupted weekly treatment at 12 mg/kg Sym004, trough levels revealed a continuous drug exposure.

A significant down-modulation of EGFR staining was observed in 17 of 18 patients in skin biopsies obtained at baseline and before the planned fifth Sym004 infusion (Fig. 3). Similar results were obtained in paired tumor specimens, which showed a significant down-modulation of EGFR staining in 9 of 11 patients (Fig. 3).

Discussion

We report results from a clinical trial in patients with SCCHN investigating the anti-tumor efficacy of Sym004, a first-in-class mixture of two chimeric mAbs targeting non-overlapping epitopes on domain III of EGFR. Our results indicate modest anti-tumor activity of Sym004 in patients with extensive pretreatment for advanced SCCHN including progression upon anti-EGFR mAbs.

According to the descriptive statistics applied to evaluate the primary endpoint, the proportion of patients alive and progression free at 6 months was 12 %, and the median PFS interval was 82 days, indicating anti-tumor activity in extensively pretreated patients with SCCHN and after exposure to prior anti-EGFR mAb treatment. The broad CIs for both measures, however, limit any further firm interpretations beyond the latter. Nevertheless, it is remarkable that at the end of the available treatment spectrum for the patients enrolled in this trial, the PFS and OS measures were numerically in the range of activity seen with zalutumumab in a phase 3 trial in patients not pre-exposed to anti-EGFR mAb treatment [5], even though the comparison itself must remain speculative. Therefore, we consider the disease stabilization with actual tumor shrinkage seen in a third of patients (Fig. 2) as a clinically notable anti-tumor efficacy, especially since our clinical findings are in line with the biological hypothesis described in preclinical models [16].

The pronounced down-regulation of EGFR in skin and tumor biopsies (Fig. 3) further supports the biological hypothesis for the proposed mechanism of action of Sym004, even though the small sample size precludes any qualitative and quantitative correlation with anti-tumor activity of Sym004. The proof of concept findings of pharmacodynamic changes allow to speculate that consequent receptor down-regulation overcomes some of the molecular changes associated with resistance to anti-EGFR blockade and in particular those that still involve EGFR, such as increased levels of high-affinity ligands and EGFR crosstalk with other receptor tyrosine kinases [7, 9, 10].

The safety profile of Sym004 was consistent with that of available anti-EGFR mAbs. However, the grade ≥3 skin toxicities (50 % grade 3 events) and hypomagnesemia (38 % grade ≥3 events) seen with Sym004 appeared to be more frequent than comparable events seen with cetuximab, panitumumab or zalutumumab in anti-EGFRmAb-naïve patients with SCCHN, with ranges of 9–22 and 4–12 %, respectively [2, 5, 6]. The reason for the observed grade ≥3 event rate may be found in the documented pronounced EGFR down-modulation by Sym004 and/or the re-exposure to anti-EGFR mAb treatment. Also, a propensity to develop high-grade skin toxicities might be explained by

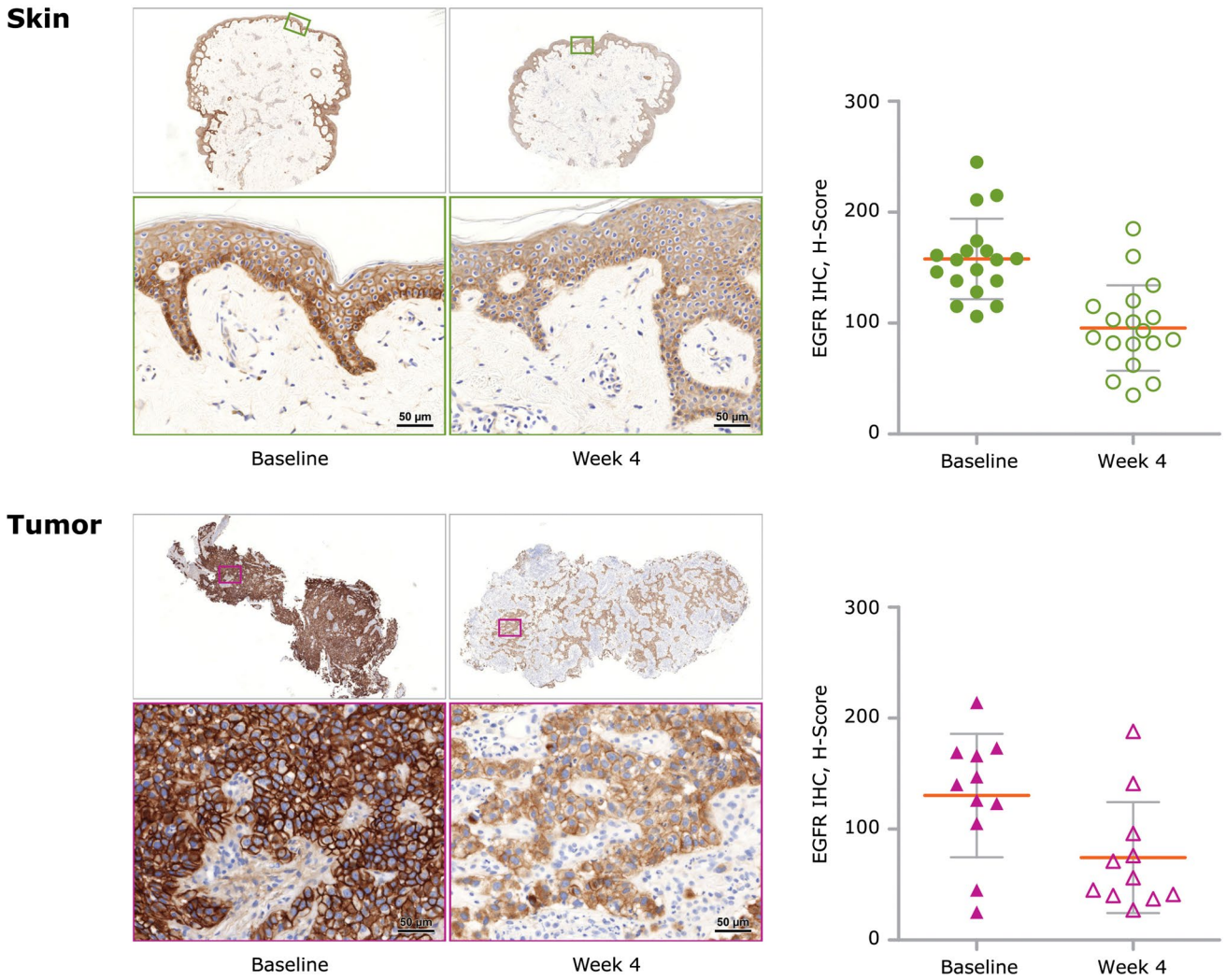

Fig. 3 Representative examples of cutaneous (*upper left panel*) and tumor (*lower left panel*) membrane EGFR down-modulation from baseline after 4 weeks of Sym004 treatment. The tissues were stained and evaluated for EGFR expression as described in the "Methods" section. Significant differences in membrane EGFR histoscores (H-score) for skin (*upper right plot*) and tumor biopsies (*lower right plot*) obtained at baseline and prior to the planned fifth infusion of Sym004. Down-regulation was seen in 17 of 18 paired skin biopsy samples ($p < 0.0001$; Wilcoxon matched pairs test, two-tailed) and in 9 of 11 paired tumor biopsy samples ($p = 0.0098$). *Horizontal bars* represent mean (*orange bars*) and standard deviation (*gray bars*)

the inclusion of patients who had previously had a clinical benefit from anti-EGFR mAbs, which is typically linked to an experience of high-grade skin toxicity. Overall, the treatment-related toxicity remained manageable by clinical supportive care measures including dose interruptions and reductions. Most notably, no cardiac symptoms were assessed in patients with grade ≥ 3 hypomagnesemia.

We therefore conclude that Sym004 demonstrated modest anti-tumor activity in an extensively pretreated population and our trial provided proof of concept pharmacodynamic and pharmacokinetic results. The degree of anti-tumor activity in patients already pre-exposed to anti-EGFR mAb treatment and standard-of-care therapy may warrant further development of Sym004 in SCCHN.

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Conflict of interest J.-P. Machiels has advisory board function in MSD (with financial compensation) and has received research grants from Novartis, Bayer and Sanofi. A. Dietz has advisory board function in Merck Serono, Boehringer Ingelheim and Merck MSD and has received speakers honorarium from Merck Serono. T.C. Gauler has advisory board function in Boehringer Ingelheim, Merck Serono, Novartis and Symphogen, has received refunds for presentations and travel from Boehringer Ingelheim and Merck Serono and performs scientific investigations in cooperation with and with financial support by Boehringer Ingelheim and Merck Serono. N.J. Skartved, I.D.

Horak, P. Pamperin and S. Braun are employees of and have ownership interests (including warrants) in Symphogen A/S. All remaining authors have declared no conflicts of interest.

Ethical standard All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards. This article does not contain any studies with animals performed by any of the authors.

Informed consent Informed consent was obtained from all individual participants included in the study.

References

1. Vermorken JB, Mesia R, Rivera F et al (2008) Platinum based chemotherapy plus cetuximab in head and neck cancer. N Eng J Med 359:1116–1127
2. Vermorken JB, Stöhlmacher-Williams J, Davidenko I et al (2013) Cisplatin and fluorouracil with or without panitumumab in patients with recurrent or metastatic squamous-cell carcinoma of the head and neck (SPECTRUM): an open-label phase 3 randomised trial. Lancet Oncol 14:697–710
3. León X, Hitt R, Constenla M et al (2005) A retrospective analysis of the outcome of patients with recurrent and/or metastatic squamous cell carcinoma of the head and neck refractory to a platinum-based chemotherapy. Clin Oncol (R Coll Radiol) 17:418–424
4. Stewart JS, Cohen EE, Licitra L et al (2009) Phase III study of gefitinib compared with intravenous methotrexate for recurrent squamous cell carcinoma of the head and neck. J Clin Oncol 27:1864–1871 **Erratum in: J Clin Oncol 2009;27:3410**
5. Machiels JP, Subramian S, Ruzsa A et al (2011) Zalutumumab plus best supportive care versus best supportive care alone in patients with recurrent or metastatic squamous-cell carcinoma of the head and neck after failure of platinum-based chemotherapy: an open-label, randomised phase 3 trial. Lancet Oncol 12:333–343
6. Vermorken JB, Herbst RS, Leon X, Amellal N, Baselga J (2008) Overview of the efficacy of cetuximab in recurrent and/or metastatic squamous cell carcinoma of the head and neck in patients who previously failed platinum-based therapies. Cancer 112:2710–2719
7. Brand TM, Iida M, Stein AP et al (2014) AXL mediates resistance to cetuximab therapy. Cancer Res 74:5152–5164
8. Tinhofer I, Klinghammer K, Weichert W et al (2011) Expression of amphiregulin and EGFRvIII affect outcome of patients with squamous cell carcinoma of the head and neck receiving cetuximab–docetaxel treatment. Clin Cancer Res 17:5197–5204
9. Wheeler DL, Huang S, Kruser TJ et al (2008) Mechanisms of acquired resistance to cetuximab: role of HER (ErbB) family members. Oncogene 27:3944–3956
10. Pedersen MW, Jacobsen HJ, Koefoed K et al (2010) Sym004: a novel synergistic anti-epidermal growth factor receptor antibody mixture with superior anticancer efficacy. Cancer Res 70:588–597
11. Skartved NJ, Jacobsen HJ, Pedersen MW et al (2011) Preclinical pharmacokinetics and safety of Sym004: a synergistic antibody mixture directed against epidermal growth factor receptor. Clin Cancer Res 17:5962–5972
12. Dienstmann R, Tolcher AW, Papadopoulos KP et al (2011) Phase I trial of the first-in-class EGFR antibody mixture, Sym004, in patients with advanced solid tumors. J Clin Oncol 2011 ASCO Annual Meeting Abstracts. vol 29, No 15_suppl (May 20 Supplement):3089
13. Dienstmann R, Tabernero J, Van Cutsem E et al (2013) Proof-of-concept study of Sym004, an anti-EGFR monoclonal antibody (mAb) mixture, in patients (pts) with anti-EGFR mab-refractory KRAS wild-type (wt) metastatic colorectal cancer (mCRC). J Clin Oncol 2013 ASCO Annual Meeting Abstracts. vol 31, No 15_suppl (May 20 Suppl):3551
14. Eisenhauer EA, Therasse P, Bogaerts J et al (2009) New response evaluation criteria in solid tumours: revised RECIST guideline (version 1.1). Eur J Cancer 45:228–247
15. Duray A, Descamps G, Bettonville M et al (2011) High-prevalence of high-risk human papillomavirus in palatine tonsils from healthy children and adults. Otolaryngol Head Neck Surg 145:230–235
16. Iida M, Brand TM, Starr MM et al (2013) Sym004, a novel EGFR antibody mixture, can overcome acquired resistance to cetuximab. Neoplasia 15:1196–1206