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The MITO CERV-2 trial: A randomized phase II study of cetuximab plus carboplatin and paclitaxel, in advanced or recurrent cervical cancer

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HIGHLIGHTS

- Cervical cancer cells often express the epidermal growth factor receptor (EGFR).
- However, the addition of cetuximab to chemotherapy did not produce benefit for advanced/recurrent cervical cancer patients.
- The presence of PIK3CA mutations may be associated with resistance to cetuximab treatment.

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ABSTRACT

Background. Cervical cancer cells often express Epidermal Growth Factor Receptor (EGFR). Cetuximab (CET), an anti-EGFR antibody, can be safely combined with carboplatin (C) and paclitaxel (P), a standard treatment for advanced/recurrent cervical cancer (ARCC) patients.

Patients and methods. ARCC patients, ECOG PS ≤ 1, were randomized to CP for 6 cycles with or without CET (400 mg/m^2 one week before starting CP, then 250 mg/m^2 weekly) until disease progression or unacceptable toxicity. Event-free survival (EFS) was the primary endpoint. With a 4.5 months expected median EFS and a 6.4 months predicted EFS (HR 0.70), 0.20 one-tailed α and 80% power, 89 events were required for the final intent-to-treat analysis.

Results. 108 patients were assigned to CP (n = 53) or CP-CET (n = 55). Median age was 50, 69% were PS0, 76% had recurrent disease, 91% had distant metastasis and 57% had received previous chemotherapy. After a median follow-up of 23 months, 102 patients had an event, 97 progressed and 61 died. Median EFS was 4.7 and 6.0 months (one-tail $P = 0.43$), median PFS was 5.2 and 7.6 months (one-tail $P = 0.20$) and median OS was 17.7 and 17 months (one-tail $P = 0.27$), with CP and CP-CET, respectively. There was no difference in the occurrence of severe adverse events, except for skin toxicity. Biomarker analysis, in a small subgroup of patients, suggests that PIK3CA mutation might be predictive of CET resistance.

Conclusion. CP-CET was not more active than CP alone in unselected ARCC patients.

1. Introduction

Cervical cancer is the fourth most common female cancer worldwide and has the fourth highest cancer-related mortality rate in women, with estimates of 528.000 new cases and 266.000 deaths from the disease per year [1]. Effective screening and prevention programs have resulted in a 75% decrease in the incidence and mortality of cervical cancer over the past 50 years. However, in developing countries, cervical cancer accounts for almost 12% of all cancers in females and remains the most common cancer in women in eastern and central Africa.

Surgery and chemo-radiotherapy may cure early-stage disease. However, patients with recurrent or metastatic cancer have limited therapeutic options. The epidermal growth factor receptor (EGFR) pathway is important in cervical cancer progression; EGFR is expressed with increasing levels from intraepithelial to localized and to invasive metastatic disease [2,3]. Cetuximab, an anti-EGFR monoclonal antibody effective in colorectal [4–6] and head and neck cancer [7], inhibits tumor proliferation in cervical tumor cell lines [3], and can be safely combined with full-dose carboplatin and paclitaxel [8], which is a standard option for treatment of advanced or recurrent cervical cancer patients [9–12].

We performed a randomized phase 2 study to measure the activity of the addition of cetuximab to carboplatin plus paclitaxel.

2. Patients and methods

2.1. Study design

A phase 2, prospective, open-label, randomized (1:1), multicenter clinical trial was performed (ClinicalTrials.gov ID: NCT00997009 and EudraCT number: 2009-010099-74), comparing Carboplatin plus Paclitaxel (standard) versus Carboplatin plus Paclitaxel plus Cetuximab (experimental).

Event-free survival (EFS) was the primary end-point, defined as the time from randomisation to progression, death without progression, premature definitive discontinuation of the whole treatment or loss to follow up, whichever occurred first.

Because of the exploratory nature of the study, a so-called relaxed statistical design was applied: one-tail test, significance level equal to 20%, power equal to 80% [13,14]. Assuming a median EFS in the standard arm equal to 4.5 months and a median EFS in the experimental arm equal to 6.4 months (corresponding to a Hazard ratio of 0.70), 89 events were required and 108 patients (54 for each arm) were planned (EAST 5 software, Cytel Software, Cambridge, MA, U.S.A.).

Secondary endpoints included progression-free survival (PFS), overall survival (OS), toxicity and objective response rate (ORR). Patient registration, randomisation (by a computer-driven minimization procedure), and data collection were performed through the website of the Clinical Trial Unit, National Cancer Institute, Napoli, Italy (http:\\www.usc-intnapoli.net). Randomisation was stratified by the number of previous chemotherapy (none vs one), the presence of distant metastases (no vs yes) and the centre. No blinding procedure was planned.

2.2. Study population

Women with advanced or recurrent cervical cancer, untreated or having failed only one previous chemotherapy (with or without concomitant or sequential radiotherapy and with at least 6 months of progression-free interval), not amenable for surgery or radiotherapy, with at least one measurable lesion by the Response Evaluation Criteria In Solid Tumors (RECIST [15]), an ECOG performance status ≤1, and a life expectancy ≥3 months were eligible. Adequate hematopoietic (absolute neutrophils count ≥1500/mm^3; platelets count ≥100,000/mm^3; hemoglobin ≥9 g/dl), hepatic (SGOT or SGPT≤3 UNL or ≤ 5 ULN in presence of liver metastases, alkaline phosphatase ≤3 ULN, total bilirubin ≤1.5 UNL), and renal (calculated creatinine clearance ≥45 ml/min) function were required.

Exclusion criteria were: a history of heart failure, angina pectoris, myocardial infarction (within 1 year from study entry), uncontrolled hypertension or arrhythmia; active infection requiring antibiotics; previous invasive malignancy within the past 5 years except non-melanoma skin cancer; residual peripheral neuropathy >grade 2 according to Common Terminology Criteria for Adverse Events (CTCAE) version 3.0; concurrent treatment with other experimental drugs; pregnancy and breast-feeding. The study was approved by Ethics Committees at each participating Institution, and the patients signed informed consent before any study related procedure.

2.3. Study treatment

Patients were randomly assigned 1:1 to receive either carboplatin plus paclitaxel (standard arm) or carboplatin plus paclitaxel plus cetuximab (experimental arm).

Patients in both arms received iv paclitaxel 175 mg/m^2 (diluted in 250 ml of 0.9% saline and infused intravenously over 3 h) and carboplatin AUC 5 (diluted in 0.9% saline solution and infused intravenously over 30 min) on day one every 21 days, for a maximum of 6 cycles.

Patients in the experimental arm received cetuximab, at a loading iv dose of 400 mg/m^2 over 120 min, one week before starting chemotherapy, and further weekly infusion at the dose of 250 mg/m^2 over 60 min, until disease progression, prolonged or unacceptable toxicity or patient's withdrawal.

In the experimental arm, a prophylactic premedication with dexamethasone 8 mg iv and antihistamine (such as diphenhydramine 50 mg) iv was mandatory before the first administration of cetuximab and strongly recommended before the following weekly doses. In both arms, a prophylactic premedication with dexamethasone 20 mg (or hydrocortisone 250 mg), chlorpheniramine 10 and ranitidine 50 mg iv was given before paclitaxel infusion.

In both arms, chemotherapy had to be deferred if absolute neutrophils count ≤1500/mm^3, platelets count ≤100,000/mm^3, or organ toxicity grade ≥ 2 (excluding alopecia and skin toxicity), for up to two consecutive weeks, until recovery. In case of longer than 2 weeks delay, chemotherapy had to be discontinued. Chemotherapy dose had to be reduced by 20% in case of neutrophils <500/mm^3 for a period of >7 days or platelets <50,000/mm^3. In case of grade ≤ 2 neurotoxicity paclitaxel dose had to be reduced by 20%, while it had to be definitively stopped in case of grade ≥ 3 neurotoxicty. In case of chemotherapy delay or stop, cetuximab could continue as planned.

Cetuximab had to be delayed in case of grade ≥ 3 skin toxicities, for up to two consecutive infusions, until toxicity resolved to grade ≤ 2. Cetuximab dose did not change after the first delay, while it had to be reduced to 200 mg/m^2 and 150 mg/m^2 after the second and third occurrence of a grade ≥ 3 skin toxicity, respectively. Dose reductions were permanent. Patients should have discontinued cetuximab if more than two consecutive infusions were withheld or on the fourth occurrence of a grade ≥ 3 skin toxicity despite appropriate dose reduction. Cetuximab infusion rate had to be decreased by 50% after the first occurrence of grade ≤ 2 allergic/hypersensitivity reaction, while it had to be definitively stopped in case of grade ≥ 3 allergic/hypersensitivity reaction or on the second occurrence of a grade ≤ 2 allergic/hypersensitivity reaction despite appropriate infusion rate reduction. In case of cetuximab delay or stop, chemotherapy could continue as planned.

2.4. Patient evaluation

Disease assessment including clinical examination, abdomino-pelvic CT scan and chest X-ray, were performed at baseline and repeated every 3 cycles during chemotherapy and every 3 months thereafter. Objective response was codified according to RECIST. Safety assessment included physical examination, blood tests (haematology and biochemistry) and collection of adverse events history, every 3 weeks during treatment and until 30 days after the last administration of study drugs. An EKG was performed every three cycles during chemotherapy. Adverse events were coded according to CTCAE version 3.0. Skin toxicity was also graded according to the MASCC (Multinational Association of Supportive Care in Cancer) EGFR-inhibitor skin toxicity tool (MESTT©) [16].

2.5. Biomarker analysis

Formalin fixed paraffin embedded (FFPE) baseline tumor samples from primary tumor or metastasis were collected for ancillary not mandatory, biomarker study. FFPE samples were analyzed at the Laboratory of Cellular Biology and Biotherapy, Istituto Nazionale per lo Studio e la Cura dei Tumori "Fondazione G.Pascale" IRCCS, Napoli- Italy by using a next generation sequencing approach based on the Ion AmpliSeq™ technology. In particular, 10 ng of genomic DNA were analyzed with the Oncomine Solid Tumor DNA kit that targets over 500 hotspot mutations in the following 22 genes: KRAS, EGFR, BRAF, PIK3CA, AKT1, ERBB2, PTEN, NRAS, STK11, MEK1, ALK, DDR2, CTNNB1, MET, TP53, SMAD4, FBXW7, FGFR3, NOTCH1, ERBB4, FGFR1, FGFR2. [17] Data analyses were performed using Ion Reporter™ Software. Mutations were confirmed by Sanger sequencing and/or Real Time PCR based assays.

2.6. Statistical analysis

Efficacy analyses were done on an intention-to-treat basis. EFS was defined as the time from randomisation to progression, death without progression, premature definitive discontinuation of the whole treatment or loss to follow-up, whichever occurred first. Patients who discontinued the treatment due to symptomatic deterioration in absence of radiologic progression were considered as progressive at the date of symptomatic deterioration. Treatment was considered as prematurely discontinued if all the planned drugs were definitively suspended for reasons different from progression or protocol completion. A patient was considered as lost to follow-up if at the date of the database lock for the primary analysis she missed the last two consecutive follow-up visits. Patients who did not have an event according to the above definition were censored at the last visit, when the patient was known to be alive and free from progression.

PFS was defined as the time from randomisation to progression or death (whichever occurred first) or date of last visit when the patient was known to be alive and free from progression. OS was defined as the time from randomisation to death or date of last follow-up for patients alive. Median follow-up was calculated according to the reverse Kaplan-Meier technique. EFS, PFS and OS curves were estimated by Kaplan-Meier product limit method and compared by log-rank test.

ORR was defined as the proportion of complete plus partial responders among patients with at least one target lesion according RECIST. Patients eligible for the evaluation of the response who did not perform the restaging were classified as "not evaluated" and conservatively included among the non-responders. Independent review of radiologic tests was not performed and no formal rules regarding blinding of local radiologists were implemented into the protocol. ORRs between the treatment arms were compared by chi-square test.

All subjects who received at least one dose of study treatment were included in compliance and safety analyses. For each toxicity and each patient, the worst degree ever suffered during treatment was used for the analysis. The whole pattern of toxicity (all grades) was considered for each item and compared between arms by exact Kruskal-Wallis linear rank test.

Statistical analysis of biomarkers data was exploratory and hypotheses generating. It was limited in its scope by the low number of samples that were collected. For the most frequent mutation (PIK3CA) the possible existence of a first-order interaction with treatment effect was tested with the likelihood-ratio test of two nested models, with and without interaction; results were summarized in a forest plot reporting HRs and 95% CIs.

Statistical analyses were performed using Stata/MP 14.2 (StataCorp LLC, College Station, TX, USA). Exact tests were performed using Cytel Studio 10 (Cytel Software, Cambridge, MA, USA).

3. Results

3.1. Patient characteristics

From February 3, 2010 to May 8, 2013, 108 patients were randomly assigned to standard (n = 53) or experimental (n = 55) arm (Fig. S1). One patient withdrew consent immediately after randomisation. Therefore, analyses included 107 patients. Baseline characteristics were balanced between the arms (Table 1). All 107 patients received at least one dose of the assigned treatment and were included in compliance and safety analyses.

3.2. Treatment compliance

Median number of chemotherapy cycles was 6 (IQR 4–6) in the both arms. Median relative dose intensity (RDI) for carboplatin was 92% (IQR 86%–100%) and 91% (IQR 85%–98%) in the standard and experimental arm, respectively. Median RDI for paclitaxel was 90% (IQR 80%–98%)

Table 1
Baseline characteristics.

	Carboplatin/paclitaxel (n = 52)	Carboplatin/paclitaxel + Cetuximab (n = 55)
Median age (IQR)	52 (44–62)	47 (41–60)
Previous chemotherapy		
No	22 (42%)	24 (44%)
Yes	30 (58%)	31 (56%)
Previous RT		
No	23 (44%)	24 (44%)
Yes	29 (56%)	31 (56%)
Previous surgery		
No	17 (33%)	14 (25%)
Yes	35 (67%)	41 (75%)
Distant metastasis		
No	9 (17%)	10 (18%)
Yes	43 (83%)	45 (81%)
ECOG performance status		
0	33 (64%)	40 (73%)
1	19 (36%)	15 (27%)
Histotype		
Adenocarcinoma	11 (21%)	12 (22%)
Squamous	41 (79%)	43(78%)
Grade		
1	1 (2%)	1 (2%)
2	8 (15%)	20 (36%)
3	31 (60%)	27 (49%)
Not known	12 (23%)	7 (13%)

and 89% (IQR 80%–94%) in the standard and experimental arm, respectively. Median RDI for cetuximab was 82% (IQR 76%–90%). At least one dose reduction was applied to chemotherapy in 17 (33%) and 21 (38%) patients in the standard and experimental arm, respectively. Cetuximab dose was reduced in 4 patients (7%). Overall, 35 (67%) and 34 (62%) patients completed the planned chemotherapy. Chemotherapy was discontinued for reasons other than completion or progression/death in 3 (6%) and 7 (13%) patients in the standard and experimental arm, respectively. Cetuximab was discontinued due to toxicity or refusal by 13 patients (24%); one patient was still on treatment at the time of the analysis (Table S1).

3.3. Activity analysis

After a median follow-up of 23 months (95% CI: 20–26), 102 patients (48 in the standard and 54 in the experimental arm) had an event for the primary analysis. The event was progressive disease for 83 patients, death without evidence of progression for 2 patients, definitive stop of the treatment for 15 patients and loss to follow-up for 2 patients. Overall, 97 patients progressed (45 in the standard and 52 in the experimental arm) and 61 died (30 and 31, respectively).

Median EFS was 4.7 (95%CI: 4.1–6.5) with the standard and 6.0 (95% CI: 4.4–7.6) months with the experimental treatment; the difference was not statistically significant (HR 0.97, 95%CI: 0.66–1.43; one-tail $P = 0.43$). Median PFS was 5.2 (95%CI: 4.5–7.8) and 7.6 (95%CI: 5.6–9.0) months (HR 0.84, 95%CI: 0.56–1.26; one-tail $P = 0.20$) and median OS was 17.7 (95%CI: 11.0–31.6) and 17 (95%CI: 12.5-NA) months (HR 0.85, 95%CI: 0.52–1.42; one-tail $P = 0.27$), with standard and experimental treatment, respectively (Fig. 1).

Twenty-one patients (8 in the standard arm and 13 in the standard arm) did actually not have measurable disease according to RECIST at baseline assessment. Therefore, 86 (80.4%) patients were eligible for response analysis, 44 (84.6%) and 42 (76.4%) in the standard and experimental arm, respectively. Nineteen patients (43%, 95%CI: 30%–58%) in the standard and 16 patients (38%, 95% CI: 25%–53%) in the experimental arm achieved an objective response ($P = 0.79$) (Table S2).

3.4. Safety analysis

One patient died for a stroke during standard treatment. There was no difference between treatment arms in the occurrence of adverse events, except, as expected, for diarrhoea (mainly grade 1) and skin toxicity (Table S3).

At least one severe (grade ≥ 3) adverse event was reported in 30 (58%) patients in the standard and 44 (80%) in the experimental arm. Severe skin toxicity was reported only in the experimental arm (9 patients reported a grade 3 skin toxicity, 6 of whom had acneiform skin rash). Twelve patients reported a severe (all grade 3) toxicity with MESTT in the experimental arm (Table S4).

3.5. Biomarker analysis

Tumor samples were collected for 20 patients, 10 in the standard and 10 in the experimental arm. Mutations were found for 5 genes out of 22 tested: PIK3CA (11 cases), KRAS (4 cases), TP53 (3 cases), ERBB2 and SMAD4 (1 case each – Fig. S2 and Table S5).

A statistically significant interaction between treatment effect on EFS and PIK3CA mutational status was found suggesting a benefit of cetuximab in the PIK3CA wild-type subgroup (HR 0.09, 95% CI: 0.01–0.87) but not in the PIK3CA mutant cohort (HR 1.69, 95% CI: 0.46–6.47; P for interaction = 0.001, Fig. S3).

4. Discussion

The addition of cetuximab to carboplatin and paclitaxel in the treatment of patients with advanced or recurrent cervical cancer, did not improve EFS, PFS, OS nor ORR in the MITO CERV-2 trial.

Other three unsuccessful phase 2 trials of cetuximab in advanced cervical cancer have been conducted. Cetuximab alone produced no objective response and an unsatisfactory PFS rate at 6 month among 35 pretreated patients, in a single-arm GOG trial [18]. A single-arm trial performed by GINECO testing the combination of cetuximab with cisplatin and topotecan as first-line chemotherapy for advanced disease was stopped early, with 19 enrolled out of 44 planned patients, for an unexpected excess of severe and fatal toxicity (infections and myelotoxicity) [19]. Finally, the combination of cetuximab with cisplatin in first line treatment was tested by GOG in 76 patients and was not active as predicted. In this study, a correlation was found between a high tumor EGFR expression and advanced stage and shorter PFS [20].

The combination tested in MITO CERV-2 (cetuximab plus carboplatin and paclitaxel) has been largely evaluated in lung cancer treatment, and was therefore considered safe. Indeed, we had no problem of tolerability and the addition of cetuximab increased skin toxicity only.

In our study, EFS was chosen as primary end-point since it includes discontinuation of treatment due to causes different from progression, such as toxicity or patient choice, as event; this might be relevant for a treatment scheduled until progression of disease. However, the addition of cetuximab did not substantially improve also the secondary outcomes.

Our study enrolled molecularly unselected patients, while genetic alterations of signalling pathways downstream the EGFR, including RAS, BRAF and PIK3CA, have been demonstrated to produce resistance to anti-EGFR agents in pre-clinical and clinical studies [21,22]. Unfortunately, collection of samples for biomarker analyses was not mandatory in our study and even if we spent a very long time trying to collect such samples we were able to collect them for <20% of the patients. However, even with limitations due to small numbers, our findings support that PIK3CA mutations might be predictive of resistance to cetuximab treatment. Our data are consistent with those coming from a randomized study of radio-chemotherapy plus cetuximab in 78 patients with locally advanced cervical cancer,

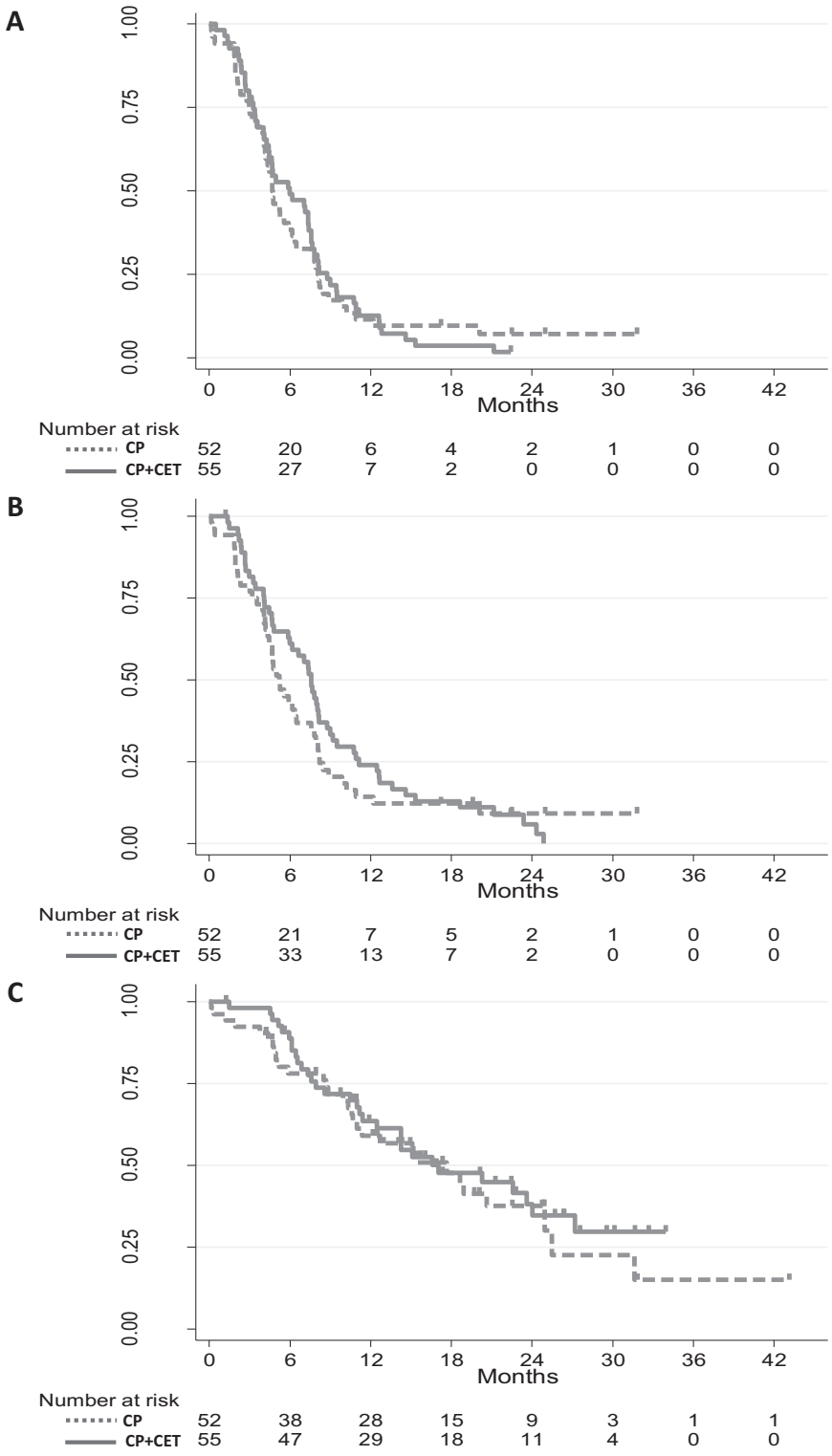

Fig. 1. Panel A: Event-free survival (EFS) curves by treatment arm. Panel B: Progression-free survival (PFS) curves by treatment arm. Panel C: Overall survival (OS) curves by treatment arm.

where no tumor had an EGFR mutation, 4% of tumors only had a KRAS mutation while 22% had a PIK3CA mutation [23]. A complete response was observed in 27% of patients without PIK3CA mutations but in none of the patient with one or more PIK3CA mutations, suggesting a correlation between these mutations and cetuximab resistance.

In conclusion, our trial showed that the addition of cetuximab to carboplatin and paclitaxel was feasible but not more effective than chemotherapy alone and does not deserve phase 3 testing in unselected advanced or refractory cervical cancer patients.

Disclosures

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All remaining authors have declared no conflicts of interest.

Authors' contribution

- Sandro Pignata, Gennaro Daniele, Francesco Perrone, Ciro Gallo, and Maria Carmela Piccirillo planned the study, pooled the data and produced initial draft.
- Simona Signoriello and Ciro Gallo performed statistical analysis.
- Sandro Pignata, Giovanni Scambia, Domenica Lorusso, Ugo De Giorgi, Maria Ornella Nicoletto, Rossella Lauria, Anna Maria Mosconi, Cosimo Sacco, Claudia Omarini, Pierosandro Tagliaferri, Gabriella Ferrandina, Saverio Cinieri, Antonella Savarese, Giorgio Valabrega, Carmela Pisano, Vanda Salutari, Francesco Raspagliesi, Barbara Kopf, Sabrina Chiara Cecere, Giulia Amadio, Giuseppa Maltese, Marilena Di Napoli, Stefano Greggi actively enrolled patients.
- Alessandra Sacco, Simona Losito and Nicola Normanno performed NGS.
- All the authors read and approved the final article.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ygyno.2019.03.260.

References

[1] Ferlay J, Soerjomataram I, Dikshit R, Eser S, Mathers C, Rebelo M, et al. Cancer incidence and mortality worldwide: sources, methods and major patterns in GLOBOCAN 2012. Int. J. Cancer. 2015;136:E359–86.
[2] Fuchs I, Vorsteher N, Buhler H, Evers K, Sehouli J, Schaller G, et al. The prognostic significance of human epidermal growth factor receptor correlations in squamous cell cervical carcinoma. Anticancer Res.. 2007;27:959–63.
[3] Bellone S, Frera G, Landolfi G, Romani C, Bandiera E, Tognon G, et al. Overexpression of epidermal growth factor type-1 receptor (EGF-R1) in cervical cancer: implications for Cetuximab-mediated therapy in recurrent/metastatic disease. Gynecol. Oncol.. 2007;106:513–20.
[4] Jonker DJ, O'Callaghan CJ, Karapetis CS, Zalcberg JR, Tu D, Au HJ, et al. Cetuximab for the treatment of colorectal cancer. N. Engl. J. Med.. 2007;357:2040–8.
[5] Van Cutsem E, Kohne CH, Lang I, Folprecht G, Nowacki MP, Cascinu S, et al. Cetuximab plus irinotecan, fluorouracil, and leucovorin as first-line treatment for metastatic colorectal cancer: updated analysis of overall survival according to tumor KRAS and BRAF mutation status. J. Clin. Oncol. Off. J. Am. Soc. Clin. Oncol.. 2011;29:2011–9.
[6] Heinemann V, von Weikersthal LF, Decker T, Kiani A, Vehling-Kaiser U, Al-Batran SE, et al. FOLFIRI plus cetuximab versus FOLFIRI plus bevacizumab as first-line treatment for patients with metastatic colorectal cancer (FIRE-3): a randomised, open-label, phase 3 trial. Lancet Oncol.. 2014;15:1065–75.
[7] Vermorken JB, Mesia R, Rivera F, Remenar E, Kawecki A, Rottey S, et al. Platinum-based chemotherapy plus cetuximab in head and neck cancer. N. Engl. J. Med.. 2008;359:1116–27.
[8] Thienelt CD, Bunn PA, Jr., Hanna N, Rosenberg A, Needle MN, Long ME, et al. Multicenter phase I/II study of cetuximab with paclitaxel and carboplatin in untreated patients with stage IV non-small-cell lung cancer. J. Clin. Oncol. Off. J. Am. Soc. Clin. Oncol.. 2005;23:8786–93.
[9] A.S. Sit, J.L. Kelley, H.H. Gallion, A.J. Kunschner, R.P. Edwards, Paclitaxel and carboplatin for recurrent or persistent cancer of the cervix, Cancer Investig. 22 (2004) 368–373.
[10] Kitagawa R, Katsumata N, Shibata T, Kamura T, Kasamatsu T, Nakanishi T, et al. Paclitaxel plus carboplatin versus paclitaxel plus cisplatin in metastatic or recurrent cervical cancer: the open-label randomized phase III trial JCOG0505. J. Clin. Oncol. Off. J. Am. Soc. Clin. Oncol.. 2015;33:2129–35.
[11] A.V. Tinker, K. Bhagat, K.D. Swenerton, P.J. Hoskins, Carboplatin and paclitaxel for advanced and recurrent cervical carcinoma: the British Columbia Cancer Agency experience, Gynecol. Oncol. 98 (2005) 54–58.
[12] H.J. Long 3rd, Management of metastatic cervical cancer: review of the literature, J. Clin. Oncol. Off. J. Am. Soc. Clin. Oncol. 25 (2007) 2966–2974.
[13] E.L. Korn, S.G. Arbuck, J.M. Pluda, R. Simon, R.S. Kaplan, M.C. Christian, Clinical trial designs for cytostatic agents: are new approaches needed? J. Clin. Oncol. Off. J. Am. Soc. Clin. Oncol. 19 (2001) 265–272.
[14] L.V. Rubinstein, E.L. Korn, B. Freidlin, S. Hunsberger, S.P. Ivy, M.A. Smith, Design issues of randomized phase II trials and a proposal for phase II screening trials, J. Clin. Oncol. Off. J. Am. Soc. Clin. Oncol. 23 (2005) 7199–7206.
[15] Therasse P, Arbuck SG, Eisenhauer EA, Wanders J, Kaplan RS, Rubinstein L, et al. New guidelines to evaluate the response to treatment in solid tumors. European Organization for Research and Treatment of Cancer, National Cancer Institute of the United States, National Cancer Institute of Canada. J. Natl. Cancer Inst.. 2000;92:205–16.
[16] M.E. Lacouture, M.L. Maitland, S. Segaert, A. Setser, R. Baran, L.P. Fox, et al., A proposed EGFR inhibitor dermatologic adverse event-specific grading scale from the MASCC skin toxicity study group, Support. Care Cancer 18 (2010) 509–522.
[17] Ojesina AI, Lichtenstein L, Freeman SS, Pedamallu CS, Imaz-Rosshandler I, Pugh TJ, et al. Landscape of genomic alterations in cervical carcinomas. Nature. 2014;506:371–5.
[18] Santin AD, Sill MW, McMeekin DS, Leitao MM, Jr., Brown J, Sutton GP, et al. Phase II trial of cetuximab in the treatment of persistent or recurrent squamous or non-squamous cell carcinoma of the cervix: a gynecologic oncology group study. Gynecol. Oncol.. 2011;122:495–500.
[19] Kurtz JE, Hardy-Bessard AC, Deslandres M, Lavau-Denes S, Largillier R, Roemer-Becuwe C, et al. Cetuximab, topotecan and cisplatin for the treatment of advanced cervical cancer: a phase II GINECO trial. Gynecol. Oncol.. 2009;113:16–20.
[20] Farley J, Sill MW, Birrer M, Walker J, Schilder RJ, Thigpen JT, et al. Phase II study of cisplatin plus cetuximab in advanced, recurrent, and previously treated cancers of the cervix and evaluation of epidermal growth factor receptor immunohistochemical expression: a Gynecologic Oncology Group study. Gynecol. Oncol.. 2011;121:303–8.
[21] Braig F, Voigtlaender M, Schieferdecker A, Busch CJ, Laban S, Grob T, et al. Liquid biopsy monitoring uncovers acquired RAS-mediated resistance to cetuximab in a substantial proportion of patients with head and neck squamous cell carcinoma. Oncotarget. 2016.
[22] Khambata-Ford S, Harbison CT, Hart LL, Awad M, Xu LA, Horak CE, et al. Analysis of potential predictive markers of cetuximab benefit in BMS099, a phase III study of cetuximab and first-line taxane/carboplatin in advanced non-small-cell lung cancer. J. Clin. Oncol. Off. J. Am. Soc. Clin. Oncol.. 2010;28:918–27.
[23] A. de la Rochefordiere, M. Kamal, A. Floquet, L. Thomas, P. Petrow, T. Petit, et al., PIK3CA pathway mutations predictive of poor response following standard radiochemotherapy +/− Cetuximab in cervical cancer patients, Clin. Cancer Res. 21 (2015) 2530–2537.