Association between neutropenia and response to ramucirumab and paclitaxel in patients with metastatic gastric cancer

Running title: **Ramucirumab-related Neutropenia and response**

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Abstract

Purpose. The aim of this study was to retrospectively evaluate if the occurrence of neutropenia is correlated with response to second-line treatment with ramucirumab plus paclitaxel for metastatic gastric cancer.
Methods. This is a retrospective study of patients treated with ramucirumab plus paclitaxel.
Results. Fifty-three patients were retrospectively evaluated. Among these, 10 patients (26,5%) developed grade ≥3 neutropenia during treatment but showed better outcome compared to those reported lower grade events. Patients with grade ≥3 neutropenia reported a progression-free survival (PFS) of 6.6 months (95% CI 3.3-8.4) and overall survival (OS) of 11 months (95% CI 5.9-13.1) versus 4.4 months (95% CI 3.9-5.2) PFS and 8.7 months (95% CI 7.8-10.1) OS respectively in patients' group with lower grade events.
Conclusions. Despite the small number of patients and the retrospective nature of the data, our analysis showed that the occurrence of neutropenia predicts response to treatment with ramucirumab and paclitaxel in patients with metastatic gastric cancer.

Keywords: gastric cancer, ramucirumab, neutropenia

Introduction

Ramucirumab is a human IgG 1 monoclonal antibody against vascular endothelial growth factor receptor 2 (VEGFR2) that prevents ligand binding and receptor-mediated pathway activation in endothelial cells, resulting in the inhibition of angiogenesis **[1]**. Two-phase III clinical trials have shown that ramucirumab is effective and safe in metastatic gastric cancer **[2,3]**. In 2014, the REGARD trial showed that ramucirumab reported a survival benefit of about 2 months against placebo **[2]**. In addition, the RAINBOW trial investigated ramucirumab or placebo plus paclitaxel in patients with advanced gastric cancer **[3]**, reporting an increased overall survival (OS) in the ramucirumab plus paclitaxel group compared to the placebo plus paclitaxel group (median 9.6 months versus 7.4 months respectively; hazard ratio (HR)=0.81). Among secondary endpoints, the progression-free survival (PFS) was longer in the experimental arm (2.7 versus 4.4 months; HR = 0.63) and the response rate (RR) was 28% for ramucirumab plus paclitaxel versus 16% for the control arm. Based on these results, ramucirumab monotherapy or in combination with paclitaxel is widely considered the standard option for patients with metastatic gastric cancer progressed after a first-line of chemotherapy. Additional confirmation of ramucirumab validity for gastric cancer comes from the RAMoss study that evaluated the safety and efficacy profile of ramucirumab in the “real-life setting” from 25 Italian hospitals recruiting more than 150 patients **[4]**. After a median follow-up of 11 months, median OS was 8.3 months and median PFS was 4.5 months for patients treated with ramucirumab plus paclitaxel.

Recently, an exposure-efficacy analysis of ramucirumab from the REGARD and RAINBOW trials suggested a positive relationship between efficacy and ramucirumab

exposure. In RAINBOW trial, grade ≥3 adverse events such as hypertension, leukopenia, and neutropenia significantly correlated with ramucirumab minimum trough concentration at steady state ($C_{min,ss}$), that was a significant predictor of OS and PFS **[5]**, suggesting that grade ≥3 neutropenia should be a predictor of efficacy in patients treated with ramucirumab plus paclitaxel. Therefore, the aim of this study is to evaluate whether the development of grade ≥3 neutropenia in patients with metastatic gastric cancer receiving ramucirumab plus paclitaxel is associated with the antitumor effect of the drug.

Patients and Methods

We performed a retrospective study conducted in 25 different oncological centres. The main methodology has already been reported **[6,7]** and here summarized for convenience. Only patients with histologically proven advanced gastric cancer progressed after standard first-line chemotherapy (doublet or triplet) were evaluated. All other eligibility criteria fit with the standard indications for ramucirumab-based chemotherapy. Patients with an operable disease or medical contraindication for an anti-angiogenetic therapy (e.g., poorly controlled hypertension, gastrointestinal perforation, fistulae or recent arterial thromboembolic event) were excluded. All patients provided written informed consent prior any medical treatment.

The clinical, radiological and biochemical pre-treatment evaluations were performed within 2 weeks the treatment beginning; for adverse events, including neutropenia, National Cancer Institute - Common Toxicity Criteria toxicity scale V.4.2 were used **[8].** Tumour response was assessed using RECIST 1.1 criteria **[9]**.

Standard schedule of ramucirumab plus paclitaxel has been administered until disease progression, unacceptable toxicity or consent withdrawal.

The aim of this study was to evaluate if the development of a grade ≥3 neutropenia correlates with efficacy and survival of patients treated with ramucirumab plus paclitaxel in a second-line regimen. For this purpose, patients were divided into two groups according to the development of grade ≥3 neutropenia as a cut-off. The primary endpoint was the PFS whereas secondary endpoints included OS and disease control rate (DCR). Kaplan-Meier method with log-rank test were performed to analyse PFS and OS in relation to the development of grade ≥3 neutropenia.

Statistical analysis was performed using STATA software with a statistical significance threshold agreed upon a $P<0.05$.

Results

Patient characteristics

The original database of patients treated from October 2015 to November 2017 was updated with the inclusion of patients treated up to January 2019, for a total of 53 patients evaluated. Baseline patient characteristics are summarised in **table 1**. The vast majority of patients were males (37; 69.8%), with a median age of 66 years (range 38–77). Overall, 22 (41.5%) patients had an ECOG performance status of 0, 20 (37.7%) patients received primary tumour resection surgery, 18 (33.9%) patients had ≥3 sites of metastasis and 20 (37.7%) patients presented peritoneal metastases.

Median PFS was 4.6 months (95% CI 4-6.1) and median OS was 8.9 months (95% CI 8-11); no complete responses (CR) were observed and DCR was 75.5% (40/53 patients) (**table 2**).

Neutropenia and clinical outcome

Ten patients (26.5%) developed grade ≥3 neutropenia during treatment (1 patient reported grade 4 neutropenia); however, none of the patients discontinued ramucirumab plus paclitaxel because of neutropenia occurrence. Among these 10 patients with grade ≥3 neutropenia, treatment delay occurred in 4 (40%) patients, with an average delay of 7 days. A dose reduction of paclitaxel was required for 3

(30%) patients. No febrile neutropenia occurred and no granulocyte colony-stimulating factor (G-CSF) was used to treat neutropenia.

Patients who developed grade ≥3 neutropenia had a median PFS of 6.6 months (95% CI 3.3-8.4) in comparison to the PFS of 4.4 months (95% CI 3.9-5.2) for patients with grade <3 neutropenia (p=0.02) (**Figure 1**). Patients with grade ≥3 neutropenia had a median OS of 11.9 months (95% CI 5.9-13.1) while, in contrast, patients with grade <3 neutropenia reported and OS of 8.7 months (95% CI 7.8-10.1) (p=0.04) **(Figure 2)**. DCR was achieved by 90% of patients with grade ≥3 neutropenia and by 72.1% of patients with no severe neutropenia (p=0.38) **(Table 2).**

After adjusting for clinical covariates (peritoneal metastases, ECOG PS, number of metastatic sites, presence of a primary tumour, time-to-progression since prior therapy, tumour differentiation grade), grade ≥3 neutropenia showed an hazard ratio (HR) of 0.44 for PFS (HR=0.44, 95% CI 0.21-0.94, p= 0.03) and of 0.45 for OS (HR=0.45, 95% CI 0.20-0.99, p=0.05).

Discussion

Inhibition of angiogenesis is still one of the most important approaches in treating patients with metastatic solid tumours **[10]**. The combination of paclitaxel chemotherapy with the ramucirumab antiangiogenetic therapy is now considered the gold standard in second-line treatment for patients with metastatic gastric cancer who progressed after standard first-line chemotherapy. Neutropenia is a possible side effect during therapy with paclitaxel plus ramucirumab as reported in the RAINBOW trial, where more than the 50% of patients developed neutropenia and more than 40% of patients developed grade ≥3 neutropenia **[3].** In the RAMoss Study, grade ≥3 neutropenia was observed in 5.4% of patients who received paclitaxel plus ramucirumab. In the current study, about the 26% of patients developed grade ≥3 neutropenia and, despite the small patients' group size, these patients had a longer PFS and OS (**Figure 1,2**) than those with grade <2 neutropenia **[4]**. A recent pharmacokinetic analysis of both REGARD and RAINBOW phase-III trials showed that grade ≥3 neutropenia significantly correlated with efficacy in the 321 patients treated with ramucirumab plus paclitaxel from the RAINBOW study **[5]**. In addition, another exploratory analysis of efficacy and safety of ramucirumab, conducted in East Asian patients from the RAINBOW trial **[11],** showed that efficacy of ramucirumab plus paclitaxel is related to the development of grade ≥3 leukopenia and neutropenia, but not hypertension **[11].** These data suggest that higher grade adverse events caused by therapy are related to a higher ramucirumab exposure that may result in longer survival. Although our data seem to confirm the role of grade ≥3 neutropenia as a predictor of efficacy during paclitaxel plus ramucirumab therapy, the small sample size of our study did not allow to perform a deeper stratification of

patients according to all grades of neutropenia and, consequently, the possible role of different grades of neutropenia remains unclear.

To date, there is a strong need to discover possible predictors of ramucirumab efficacy in metastatic gastric cancer. Statistically significant prognostic factors of efficacy of ramucirumab plus paclitaxel in the analysis of REGARD and RAINBOW trials and other retrospective studies are reported in **Tables 3 and 4** respectively. In 2017, a pooled analysis from the REGARD and RAINBOW phase III trials evaluated more than 1000 patients in order to discover baseline prognostic factors of patients' survival treated with ramucirumab **[12]**. A total of 41 baseline clinical and laboratory factors were analysed; the authors found 12 prognostic factors of poor survival **(Table 3).** In addition, data from baseline prognostic quality of life parameters showed that appetite loss is also an independent prognostic factor. In the RAMoss study, the presence of peritoneal metastases and ECOG performance status were independent poor prognostic factors **[4] (Table 4)**. In 2018, two small studies showed that hypertension may be predictive of better outcomes in patients affected by gastric cancer who were treated with paclitaxel plus ramucirumab **[6,13] (Table 4)**. However, data on the role of hypertension as a side effect related to efficacy of an antiangiogenic drug is well established in other several tumours in addition to gastric cancer **[14-17].** Recently, a multicenter retrospective study aimed to evaluate the correlations between Body Weight Loss, Body Mass Index and clinical outcomes of metastatic gastric cancer patients treated with second-line ramucirumab-based therapy **[18].** The study showed that Body Weight Loss seems not to have correlation with clinical outcome whereas Body Mass Index and ECOG performance status remain major prognostic factors (**Table 4**). However, the authors found a possible

explanation for the lack of prognostic effect of Body Weight Loss in the proportion of patients underwent surgery (more than the half) **[18]**. Finally, two retrospective analysis confirmed the efficacy of ramucirumab in patients with ascites and across age **[19,20].** In this setting, our study provides a possible predictor of efficacy during the treatment with ramucirumab and not only a baseline clinical pathological factor as reported by the vast majority of published studies.

Our study presents several limitations that we need to report: first, the retrospective nature of the data; secondly, the small number of patients evaluated and the absence of a control arm. However, although it is difficult to draw a definitive conclusion, we observed a strong correlation between the occurrence of grade ≥3 neutropenia and the response to ramucirumab therapy.

In conclusion, ramucirumab-induced neutropenia is a predictor factor of treatment efficacy and survival in patients with metastatic gastric cancer treated with the combination of ramucirumab plus paclitaxel. Prospective large-scale trials are needed to further confirm these results.

Conflict of Interest:

All authors declare no actual or potential conflicts of interest, including any financial, personal or other relationships with people or organizations (within three years from the beginning of the submitted work) that could influence, or be perceived to influence, this work**.**

Legends

Figure 1: A) Estimated PFS for ramucirumab and paclitaxel in patients with grade ≥3 neutropenia (blue) or without (red).

Figure 2: A) Estimated OS for ramucirumab and paclitaxel in patients with grade ≥3 neutropenia (blue) or hypertension <3 (red).

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