bjh short report

The use of erythropoiesis-stimulating agents is safe and effective in the management of anaemia in myelofibrosis patients treated with ruxolitinib

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Summary

Erythropoiesis-stimulating agents (ESAs) were combined with ruxolitinib in 59 anaemic myelofibrosis patients (93% with Dynamic International Prognostic Scoring System [DIPSS] intermediate-2/high risk; 52·5% transfusion-dependent). Anaemia response (AR) rate was 54% and 76% of patients responded at 5 years. A further 15% displayed minor improvement in anaemia and 78% of patients reduced spleen size. Endogenous erythropoietin levels <125 u/l correlated with a higher AR rate (63% vs. 20%, $P = 0{\cdot}008$). No thrombotic events or other toxicities occurred. Overall survival was 62% at 4 years, influenced by DIPSS and transfusion dependency. ESAs seem effective in improving anaemia in ruxruxolitinib-treated myelofibrosis patients.

Keywords: myelofibrosis, anaemia, erythropoiesis stimulating agents, ruxolitinib, erythropoietin.

The Janus Kinase 2 (JAK2) inhibitor ruxolitinib is currently the main approved treatment for symptomatic myelofibrosis (MF) with splenomegaly and is effective in reducing spleen size, MF symptoms, and possibly prolonging survival (Harrison *et al*, 2012; Verstovsek *et al*, 2012; Cervantes & Pereira, 2017). However, consistent with its known mechanism of action, one of ruxolitinib's main side effects is anaemia, which occurs in at least 40% of patients and can be a limiting factor for treatment tolerability and optimal dosage, mostly in the first 12–24 weeks of treatment. Moreover,

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ruxolitinib has not been reported to improve the haemoglobin (Hb) level in patients already anaemic at treatment start (Harrison *et al*, 2012; Verstovsek *et al*, 2012). The use of erythropoiesis-stimulating agents (ESAs) was discouraged in the COMFORT I study, however some responses to ESAs were seen in 13 patients from the COMFORT II study and reported in a *post hoc* analysis (McMullin *et al*, 2015).

Here we present our retrospective multi-centre experience on a combination therapy with ruxolitinib and ESAs on the largest patient series published so far.

Methods

ESAs (epoetin alpha/beta/zeta or darbepoetin) were given off-label to treat anaemia (Hb <100 g/l) of all MF patients attending our institutions. Ruxolitinib was given to MF patients classified as International Prognostic Scoring System (IPSS) intermediate 2 or high for symptomatic disease or splenomegaly, according to the approved indications in Italy; a few patients with IPSS intermediate 1 MF received it in a compassionate use programme. We retrospectively evaluated 59 consecutive patients who received ruxolitinib combined with ESAs for anaemia.

Epoetin alpha/zeta, beta and darbepoetin were administered subcutaneously at the starting weekly doses of 40 000 international units (iu), 30 000 iu and 150 μg, respectively; these could be doubled in case of no response. Ruxolitinib was given at the standard dosage with adjustment in case of toxicity, according to manufacturer recommendations.

The anaemia response (AR) rate was defined according to the International Working Group-Myeloproliferative Neoplasms Research and Treatment (IWG-MRT) criteria (Tefferi *et al*, 2013). Haematological improvement was defined as transfusion decrease of >50% or sustained Hb increase of 10–20 g/l in transfusion independent patients (Cervantes *et al*, 2004).

Patients' characteristics were compared using Pearson chi-square test for the categorical variables and the Kruskall–Wallis test for the continuous ones. Overall survival (OS) and response duration were estimated by Kaplan Mayer method from ESAs start until death or last follow-up and from the achievement of AR to treatment failure, respectively; any statistical difference between curves was assessed by log rank test. Patients who discontinued ESAs still in response were censored at the time of treatment discontinuation. Univariate and multivariate analysis were performed using the Cox model. P values <0·05 were considered statistically significant.

Results

Fifty-nine patients diagnosed with MF who received ESAs together with ruxolitinib (n = 9) or after being on ruxolitinib (n = 50) for a median time of 4 months (range 1–38 months) were investigated. Median time from diagnosis to ESAs start was 19 months (range 0–81) and to ruxolitinib start 14 months (0–70). Table I lists the patients characteristics at ESAs initiation.

Forty-five patients (76%) were already anaemic (Hb <100 g/l) before starting ruxolitinib: 9/45 received ESAs together with ruxolitinib and 36/45 after a median of 5 months (range 1–38). The remaining 14 patients became anaemic after receiving ruxolitinib and subsequently started ESAs treatment.

Patients received ESAs for a median time of 15 months (range 2–55). AR rate was 54% (32/59 patients) with a median time to AR of 4 months (range 1–13 months) with 95% and 76% of the patients still responding at 4 and 5 years, respectively (Fig 1).

Similar AR rates (55% and 53%) were observed in the 29 patients who started ESAs after being on ruxolitinib for >3 months (median time for ruxolitinib-induced anaemia resolution), and 30 patients who started ESAs at the same time or within 3 months of ruxolitinib initiation.

Anaemia response seemed more frequent in the 46 patients (78%) who showed a reduction in spleen size with ruxolitinib than in non-responding patients (61% vs. 33%: P = 0·088).

Table I. Patients characteristics at the start of therapy with erythropoiesis-stimulating agents.

	n (%) or n (range)
Age, years	69 (48–81)
Gender (male/total)	35/59 (59%)
PMF	23/59 (39%)
PPV-MF	16/59 (27%)
PET-MF	20/59 (34%)
JAK2 V617F	42/50 (84%)
CALR	7/50 (14%)
MPL	1/50 (2%)
RBC transfusion dependency	31/59 (52·5%)
Haemoglobin (g/l)	87 (60–100)
Leucocyte count ($\times 10^9$/l)	9 (1–31)
Serum ferritin levels (μg/l)	344 (5–6134)
Median EPO level (u/l)	70 (2–674)
EPO serum levels <125 u/l	27/42 (64%)
DIPSS Intermediate - 1	4/59 (7%)
DIPSS Intermediate - 2	42/59 (71%)
DIPSS High	13/59 (22%)
Darboepoetin	5/59 (8%)
Erythropoietin	54/59 (92%)
Epoetin alpha	18/54 (33%)
Epoetin beta	30/54 (56%)
Epoetin zeta	6/54 (11%)

DIPSS, Dynamic International Prognostic Scoring System; EPO, erythropoietin; PET-MF, post-essential thrombocytopenia myelofibrosis; PMF, primary myelofibrosis; PPV-MF, post-polycythaemia vera myelofibrosis; RBC, red blood cells.

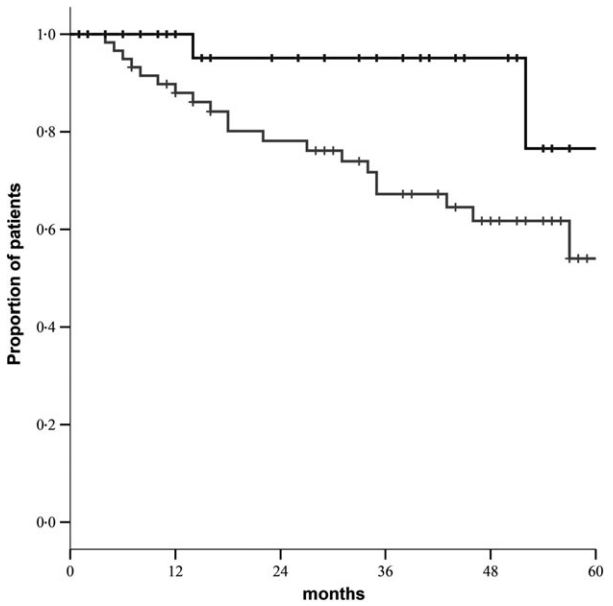

Fig 1. Duration of anaemia response (black line; median not reached) and overall survival (grey line; median 61 months) of myelofibrosis patients treated with ruxolitinib and erythropoiesis-stimulating agents.

Lower endogenous erythropoietin (EPO) levels were a significant predictor of AR, with 63% of patients with EPO <125 u/l responding vs. 20% in the EPO >125 u/l group ($P = 0{\cdot}008$). We were unable to identify any other predicting factors of response to ESAs. In particular, the Dynamic IPSS (DIPSS), serum ferritin level at baseline, blood counts, mutational status for *JAK2*, *CALR* or *MPL*, age, disease duration and transfusion dependency did not have any significant impact on response. Indeed, 61% of transfusion-dependent patients responded, as compared to 46% of transfusion-independent patients ($P = 0{\cdot}253$).

Of note, a further 15% of patients achieved an improvement in Hb levels without meeting the criteria of AR, leading to an overall response rate of 69% (41/59 patients).

No thrombotic events or toxicities**,** except for mild nausea**,** were reported during ESA treatment with. An increase in spleen size during ESA treatment was observed in only 1 case of ruxolitinib-responsive patients.

Twenty-one patients (35·5%) died. The 2- and 4-year OS from ESA start was 78% and 62% (Fig 1), respectively; at multivariate analysis, OS was significantly affected by DIPSS (hazard ratio [HR] 3·68, 95% confidence interval [CI] 1·17–1·09, $P = 0{\cdot}017$) and transfusion dependency (HR 4·00, 95% CI 1·41–11·29, $P = 0{\cdot}006$) whereas the impact of AR was border line (HR 0·43, 95% CI 0·17–1·19, $P = 0{\cdot}076$).

Discussion

Anaemia is one of the main issues in MF with an unmet need for approved drugs. Our centres have routinely employed ESAs in approximately 100 MF patients with an AR rate (49%) similar to that recently reported in a larger series (Hernández-Boluda *et al*, 2016). More recently, we have extended ESAs to anaemic MF patients receiving ruxolitinib. In Italy, ruxolitinib prescription is limited to patients with splenomegaly and Intermediate 2/High IPSS score and as our patients were also anaemic, the majority had a rather poor prognosis. Nevertheless, 54% of patients achieved an AR, as reported by others with ESAs alone in patients with less unfavourable prognostic features (Hasselbalch *et al*, 2002; Cervantes *et al*, 2004, 2006; Tsiara *et al*, 2007; Huang & Tefferi, 2009).

The use of ESAs was discouraged in the first ruxolitinib clinical trials (McMullin *et al*, 2015), because of concerns regarding its possible activation of the JAK pathway, potentially counteracting ruxolitinib-induced reduction of spleen size. For the same reason, ESAs might have been expected to be less effective in the presence of *JAK2* inhibition. However, *JAK2* is probably not completely inhibited by therapeutic concentrations of ruxolitinib, possibly because of the prolonged half-life of ESAs compared to the short half-life of ruxolitinib; moreover, ruxolitinib-induced reduction of splenomegaly and inflammatory cytokines could synergize with ESAs in improving anaemia. Indeed, the AR rate seemed higher in ruxolitinib responders than in non-responsive patients. Moreover, while previous studies reported a median AR duration of about 20 months (Hernández-Boluda *et al*, 2016), in our MF patients AR seemed to be long-lasting, with 96% of patients still responding at the median follow-up of 48 months. Finally, we observed a comparable AR in transfusion-dependent and non-dependent patients, differently from what was previously described with ESAs alone (Hernández-Boluda *et al*, 2016).

In the COMFORT studies ruxolitinib-induced anaemia resolved in the majority of patients within 12 weeks (Verstovsek *et al*, 2017) but no direct correlation between anaemia improvement and ruxolitinib was clearly reported. In our series, the majority of patients were already anaemic before starting ruxolitinib and a comparable (~50%) AR rate was observed in the group of patients starting ESAs more than 3 months after ruxolitinib (when ruxolitinib-induced anaemia was probably resolved in most cases). Therefore, these data suggest that AR in the majority of cases was correlated with ESAs treatment and not only to the spontaneous resolution of the early ruxolitinib toxicity on red cells progenitors.

Our data also confirm the endogenous EPO level as a good predictor of response, which can easily be used to select patients for ESAs treatment.

Importantly, we did not see any negative impact of ESAs on response to ruxolitinib. Indeed, 78% of our patients experienced a spleen reduction with ruxolitinib, as expected from the literature (Harrison *et al*, 2012; Verstovsek *et al*, 2012) and only one responding patient had an increase in spleen

size during ESAs treatment. Moreover, no thrombotic event was observed during ESAs treatment.

Overall survival was, at least, comparable to the one reported in the COMFORT studies (Harrison *et al*, 2016; Verstovsek *et al*, 2017). In particular, response to ESAs was associated with a trend towards better survival, as recently reported (Hernández-Boluda *et al*, 2016).

Our favourable results in terms of AR are in line with the majority of previous reports on ESAs alone; however, a further 15% of patients obtained an "anaemia improvement". Moreover, the longer response duration and the higher response rate in transfusion-dependent patients, compared to results with ESAs alone, could suggest a synergistic more than antagonistic activity of ESAs and ruxolitinib, possibly due to the reduction of splenomegaly and inflammatory symptoms.

In conclusion, ESAs seem effective in improving anaemia in MF patients treated with ruxolitinib without significant toxicities, and endogenous EPO levels could help to identify patients more likely to benefit from ESAs. This is an important finding that should be confirmed prospectively in order to include ESAs in the treatment options for MF patients.

Authorship contributions

E.C. designed research, followed the patients, analysed data and wrote the paper; D.F. designed research and wrote the paper; D.C., E.M.E., V.M., G.A.P., N.P., E.B., C.F., M.C., G.L., M.M., M.M. followed the patients and collected data. M.B. supervised the research and provided funding. All authors reviewed and approved the manuscript.

Conflict of interest disclosures

No conflict of interest to disclose.

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