Original Article

Ruxolitinib Rechallenge in Resistant or Intolerant Patients With Myelofibrosis: Frequency, Therapeutic Effects, and Impact on Outcome

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BACKGROUND: After ruxolitinib discontinuation, the outcome of patients with myelofibrosis (MF) is poor with scarce therapeutic possibilities. **METHODS:** The authors performed a subanalysis of an observational, retrospective study (RUX-MF) that included 703 MF patients treated with ruxolitinib to investigate 1) the frequency and reasons for ruxolitinib rechallenge, 2) its therapeutic effects, and 3) its impact on overall survival. **RESULTS:** A total of 219 patients (31.2%) discontinued ruxolitinib for ≥14 days and survived for ≥30 days. In 60 patients (27.4%), ruxolitinib was rechallenged for ≥14 days (RUX-again patients), whereas 159 patients (72.6%) discontinued it permanently (RUX-stop patients). The baseline characteristics of the 2 cohorts were comparable, but discontinuation due to a lack/loss of spleen response was lower in RUX-again patients ($P = .004$). In comparison with the disease status at the first ruxolitinib stop, at its restart, there was a significant increase in patients with large splenomegaly ($P < .001$) and a high Total Symptom Score (TSS; $P < .001$). During the rechallenge, 44.6% and 48.3% of the patients had spleen and symptom improvements, respectively, with a significant increase in the number of patients with a TSS reduction ($P = .01$). Although the use of a ruxolitinib dose > 10 mg twice daily predicted better spleen ($P = .05$) and symptom improvements ($P = .02$), the reasons for/duration of ruxolitinib discontinuation and the use of other therapies before rechallenge were not associated with rechallenge efficacy. At 1 and 2 years, 33.3% and 48.3% of RUX-again patients, respectively, had permanently discontinued ruxolitinib. The median overall survival was 27.9 months, and it was significantly longer for RUX-again patients ($P = .004$). **CONCLUSIONS:** Ruxolitinib rechallenge was mainly used in intolerant patients; there were clinical improvements and a possible survival advantage in many cases, but there was a substantial rate of permanent discontinuation. Ruxolitinib rechallenge should be balanced against newer therapeutic possibilities. ***Cancer* 2021;0:1-9**. © *2021 American Cancer Society*.

KEYWORDS: cancer, myelofibrosis, outcome, rechallenge, ruxolitinib.

INTRODUCTION

Ruxolitinib is the first *JAK1/JAK2* inhibitor approved for the treatment of splenomegaly and symptoms related to myelofibrosis (MF), and it has demonstrated significant efficacy in most patients with improvements in quality of life and overall survival in responding patients.[1,2] Nevertheless, some patients cannot tolerate ruxolitinib, and many

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Additional supporting information may be found in the online version of this article.

DOI: 10.1002/cncr.33541, **Received:** January 18, 2021; **Revised:** February 24, 2021; **Accepted:** March 1, 2021, **Published online** Month 00, 2021 in Wiley Online Library (wileyonlinelibrary.com)

do not achieve a response or lose it over time; this leads to ruxolitinib discontinuation in approximately 50% of patients at 3 years.[3,4] After ruxolitinib discontinuation, the outcome is poor with scarce therapeutic possibilities, including palliation, investigational agents, allogeneic stem cell transplantation (ASCT), and splenectomy.[4-7] Other *JAK* inhibitors have also been studied in MF over the last several years.[8] Among them, fedratinib has recently received US Food and Drug Administration approval for use in patients intolerant of or resistant to ruxolitinib; it has a response rate of approximately 30%, regardless of the reason for ruxolitinib discontinuation.[9] Many other new agents, alone or in combination with ruxolitinib—particularly second-generation *JAK2* inhibitors (eg, momelotinib and pacritinib),[10,11] telomerase inhibitors (eg, imetelstat),[12] BET inhibitors (eg, CPI-0610),[13] PI3/AKT inhibitors (eg, buparlisib),[14] LSD1 inhibitors (bomedemstat),[15] and BCL-2/BCL-X inhibitors (eg, navitoclax)[16]—are currently under investigation.

Beyond new drugs, a retrospective case series including 13 patients has suggested that patients may respond to a rechallenge of ruxolitinib after the drug is first stopped.[17] This therapeutic strategy may be attractive in routine clinical practice because it is simple to implement and may include the frailest patients who cannot be enrolled in investigational clinical studies. However, it is not known how frequently or for what reasons ruxolitinib rechallenge is used in real-life practice, what its clinical effects are, and whether rechallenge may affect the outcome.

To answer these questions, we performed a subanalysis of an observational, retrospective study (RUX-MF) that was promoted by the L. and A. Seràgnoli Institute of Hematology in Bologna, Italy.

MATERIALS AND METHODS

Patients and Study Design

The RUX-MF observational, retrospective study involves 703 consecutive MF patients treated with ruxolitinib at 22 academic hematology centers that are dedicated to the treatment of MF. The list of the participating centers is available in the supporting information. All centers were asked to report, in an electronic case report form, their consecutive MF patients who received ruxolitinib according to standard clinical practice. The total number of medical files was reported by each center via data input into an electronic database developed to record all study data after de-identification of the patients with alphanumeric codes to protect personal privacy. The collected data included patient demographics, comorbidities, medications, clinical/laboratory tests at diagnosis and during follow-up, ruxolitinib start and stop dates, types of MF therapies before and after ruxolitinib, duration of ruxolitinib treatment, and adverse events during the treatment. Any treatment decision was at the physician's discretion, was based on a patient's characteristics, and was independent of participation in this study. After the first data entry, the follow-up information was validated with revision of clinical data, and specific queries were addressed to the participating center in cases of inconsistent data.

In this subanalysis, we included consecutive MF patients who received a primary treatment course with ruxolitinib of at least 14 days, discontinued the drug for at least 14 days while in the chronic phase, and survived for at least 30 days after discontinuation. A total of 302 patients discontinued ruxolitinib after a median observation time of 13.9 months (range, 0.5-84.5 months). Eighty-three patients were excluded from this analysis because they discontinued ruxolitinib in the accelerated/blast phase (n = 63), survived less than 30 days after discontinuation (n = 10), or discontinued ruxolitinib for less than 14 days (n = 10). Therefore, the current analysis comprises 219 chronic-phase patients who received and stopped ruxolitinib for ≥14 days and survived for ≥30 days after discontinuation. Figure 1 reports the numbers of individuals at each stage of the study. All patients were followed until death or the data cutoff (December 1, 2020).

Definitions

Diagnoses of primary MF and post-polycythemia vera/post-essential thrombocythemia MF were made according to the 2016 World Health Organization criteria or International Working Group on Myelofibrosis Research and Treatment (IWG-MRT) criteria, respectively.[18,19] All patients who received treatment with ruxolitinib in the current analysis were in the chronic phase (peripheral and marrow blast cells < 10%). The risk category was assessed at the time when patients started on ruxolitinib according to the Dynamic International Prognostic Score System.[20] Histologic examinations were performed at local institutions; fibrosis was graded according to the European Consensus Grading System.[21] An unfavorable karyotype was categorized as previously described.[22] The diagnosis of blast phase was made according to World Health Organization criteria with a 20% bone marrow or peripheral blood blast threshold

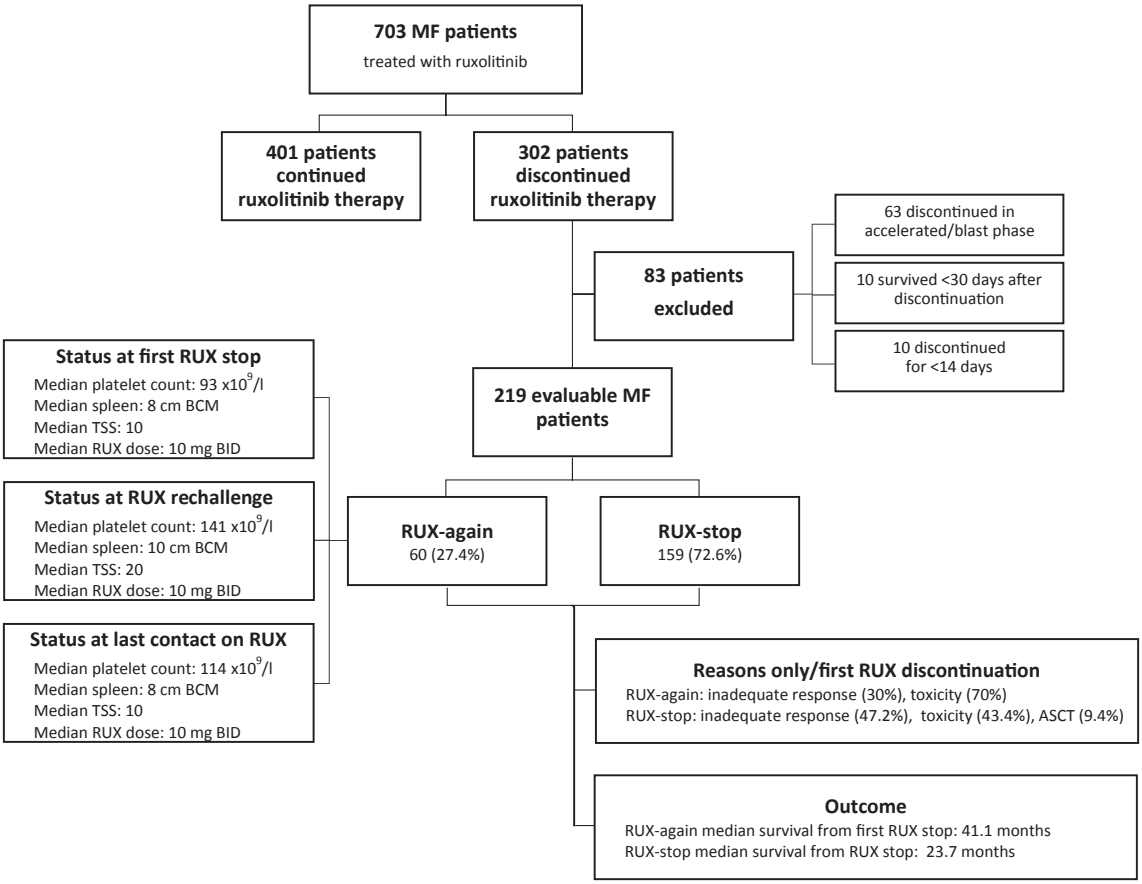

Figure 1. Study flowchart. The numbers of individuals at each stage of the study, the main descriptive results, and the major findings are reported. ASCT indicates allogeneic stem cell transplantation; BCM, below costal margin; bid, twice daily; MF, myelofibrosis; RUX, ruxolitinib; TSS, Total Symptom Score.

for diagnosis.[19] The burden of MF-related symptoms was assessed with the 10-item Myeloproliferative Neoplasm Symptom Assessment Form Total Symptom Score (TSS).[23] Spleen responses were assessed according to the 2013 IWG-MRT/European LeukemiaNet criteria.[7]

Inadequate responses included a lack of spleen response (ie, an absence of spleen response with ruxolitinib therapy for ≥3 months) and a loss of spleen response (ie, any increase in spleen size not meeting the initial response criteria at the maximum tolerated dose).[24] Notably, at the time when patients lost a spleen response, the spleen still may have been smaller than it was at the baseline.

All adverse events were defined and graded according to the Common Terminology Criteria for Adverse Events (version 4.0). Specifically, events graded as 2 or higher required active systemic treatment, and those graded as 4 were life-threatening.

Ethical Aspects

The RUX-MF study was performed in accordance with the guidelines of the institutional review boards of the participating centers and the standards of the Helsinki Declaration. The promoter of this study was the L. and A. Seràgnoli Institute of Hematology (Azienda Ospedaliera S. Orsola-Malpighi, Bologna, Italy), which obtained approval from the Area Vasta Emilia Centro ethics committee. The study was also approved by the local ethics committees of all participating centers (protocol code MF-2014-01) and had no commercial support.

Statistical Analysis

The statistical analysis was performed at the Biostatistics Laboratory of the myeloproliferative neoplasms Unit at the L. and A. Seràgnoli Institute of Hematology (IRCCS Azienda Ospedaliero–Universitaria di Bologna, Bologna, Italy). Continuous variables were summarized

as medians and ranges, and categorical variables were summarized as counts and relative frequencies (percentages) of each category. Comparisons of quantitative variables between groups of patients were performed with the Wilcoxon-Mann-Whitney rank-sum test or the Student *t* test, and associations between categorical variables (2-way tables) were tested with the Fisher exact test or the χ^2 test as appropriate. Variations in continuous and categorical variables between ruxolitinib discontinuation and rechallenge and between rechallenge and last contact on ruxolitinib were assessed with the Wilcoxon signed-rank test and the McNemar test, respectively. Treatments were considered time-to-event variables. The time to therapy was calculated from the first discontinuation of ruxolitinib to the start of the other therapy. Overall survival was estimated from the date of the first/only ruxolitinib discontinuation to last contact. Comparisons of treatments and overall survival were performed with log-rank tests. Factors associated with responses to ruxolitinib rechallenge were identified with death considered as a competing risk, according to the model of Fine and Gray, from the start of ruxolitinib rechallenge to the date of the response or last contact on ruxolitinib therapy. A multivariable analysis was not performed when ≤1 covariate had a *P* value < .10 in univariate analyses. For all tested hypotheses, 2-tailed *P* values < .05 were considered significant. Statistical analyses were performed with STATA 15.1 software (StataCorp LP, College Station, Texas).

RESULTS

Study Population

Overall, 219 of 703 total patients (31.2%) were evaluable in this study. Among these 219 patients, 60 patients (27.4%) rechallenged ruxolitinib (the RUX-again cohort), whereas 159 (72.6%) discontinued ruxolitinib permanently (the RUX-stop cohort). The main demographic, clinical, and hematological features at the start of ruxolitinib and at the first/only ruxolitinib discontinuation are presented in Table 1. No significant differences were observed between the 2 cohorts. The median duration of ruxolitinib therapy before the first/only discontinuation was 16.5 months (range, 0.5-84.5 months) and 12.3 months (range, 0.8-79.1 months) for RUX-again and RUX-stop patients, respectively (P = .41). The median follow-up after ruxolitinib discontinuation was 18.8 months (range, 1-93.7 months) for RUX-again patients and 15.5 months (range, 1.3-79.8 months) for RUX-stop patients (P = .21).

TABLE 1. Patient Characteristics

Characteristic	Study Cohort (n = 219)	RUX-Stop (n = 159)	RUX-Again (n = 60)	*P*
Age, median (range), y				
At ruxolitinib start	67.5 (24.0-88.5)	67.5 (24.0-88.5)	67.4 (40.9-87.4)	.62
At first/only ruxolitinib stop	69.3 (41.1-88.3)	68.9 (24.3-91.1)	69.3 (41.1-88.3)	.90
Male sex, No. (%)	134 (64.2)	97 (61.0)	37 (61.7)	.93
Primary MF, No. (%)	127 (58.0)	95 (59.8)	32 (53.3)	.39
DIPSS risk category at ruxolitinib start, No. (%)				
Intermediate-1	99 (45.2)	69 (43.4)	30 (50.0)	
Intermediate-2	109 (49.8)	84 (52.8)	25 (41.7)	.30
High	11 (5.0)	6 (3.8)	5 (8.3)	
Hemoglobin, median (range), g/dL				
At ruxolitinib start	10.0 (6.0-16.7)	9.9 (6.0-16.7)	10.2 (7.0-16.4)	.25
At first/only ruxolitinib stop	9.2 (5.0-15.9)	9.1 (5.7-14.3)	9.5 (5.0-15.9)	.94
Platelet count, median (range), $\times 10^9$/L				
At ruxolitinib start	217 (50-1400)	202 (55-1400)	249 (50-1026)	.44
At first/only ruxolitinib stop	111 (3-870)	114 (3-829)	93 (8-870)	.27
Leukocytes, median (range), $\times 10^9$/L				
At ruxolitinib start	9.3 (1.1-80)	8.9 (1.1-80)	12.6 (2.2-78.9)	.16
At first/only ruxolitinib stop	8.8 (1.3-118)	9.1 (1.3-118)	7 (1.7-81.7)	.23
Dose, median (range), mg bid				
At ruxolitinib start	15 (5-20)	15 (5-20)	15 (5-20)	.21
At 3 mo	10 (5-20)	10 (5-20)	15 (5-20)	.87
At first/only ruxolitinib stop	10 (2.5-25)	10 (5-20)	10 (2.5-25)	.53
Dose > 10 mg bid, No. (%)				
At ruxolitinib start	127 (58.0)	87 (54.7)	40 (66.7)	.11
At 3 mo	109 (49.8)	78 (49.1)	31 (51.7)	.85
At first/only ruxolitinib stop	86 (39.2)	64 (40.2)	22 (36.7)	.60
Total Symptom Score, median (range)				
At ruxolitinib start	20 (0-90)	20 (0-90)	20 (0-80)	.13
At first/only ruxolitinib stop	10 (0-100)	10 (0-100)	10 (0-52)	.37
Total Symptom Score ≥ 20, No. (%)				
At ruxolitinib start	74 (33.8)	59 (37.1)	15 (25.0)	.12
At first/only ruxolitinib stop	67 (30.6)	48 (30.2)	19 (31.7)	.86
Spleen size BLCM, median (range), cm				
At ruxolitinib start	12 (0-38)	11 (0-38)	12 (0-29)	.48
At first/only ruxolitinib stop	9 (0-40)	9.5 (0-40)	8 (0-28)	.56
Spleen size ≥ 10 cm BLCM, No. (%)				

TABLE 1. *Continued*

Characteristic	Study Cohort (n = 219)	RUX-Stop (n = 159)	RUX-Again (n = 60)	*P*
At ruxolitinib start	138 (63.0)	101 (63.5)	37 (61.7)	.80
At first/only ruxolitinib stop[a]	99 (46.3)	78 (50.0)	21 (36.2)	.07
Time from MF diagnosis to ruxolitinib start, median (range), mo	23.6 (0-337)	22.9 (0-317)	24.0 (0.1-337)	.80

Abbreviations: bid, twice daily; BLCM, below left costal margin; DIPSS, Dynamic International Prognostic Score System; MF, myelofibrosis; RUX-again, patients who rechallenged ruxolitinib; RUX-stop, patients who discontinued ruxolitinib permanently.
[a]Three RUX-stop patients and 2 RUX-again patients were splenectomized before ruxolitinib discontinuation.

In the 60 RUX-again patients, the main reason for temporary discontinuation was toxicity (n = 42; 70%), including grade 3 to 4 thrombocytopenia (38.1%), anemia (26.2%), infections (21.4%), and other adverse events (including second primary malignancies, liver toxicity, hemorrhages, and pleural effusions; 14.3%). In the remaining 18 patients, ruxolitinib was discontinued because of an inadequate spleen response (lack of response, 10; loss of response, 8).

Among the 159 RUX-stop patients, 75 (47.2%) discontinued ruxolitinib because of an inadequate spleen response (lack of response, 56; loss of response, 19), and 15 (9.4%) stopped while in response to undergo ASCT. Adverse events caused ruxolitinib discontinuation in the remaining 69 patients (43.4%); specifically, they were grade 3 to 4 anemia (40.6%), thrombocytopenia (27.5%), infections (18.8%), and others (including second primary malignancies and thromboses; 13.1%). Overall, the percentage of patients who discontinued because of an inadequate response was significantly higher among RUX-stop patients ($P = .004$).

Efficacy of Ruxolitinib Rechallenge

At the first ruxolitinib discontinuation, 36.2% of RUX-again patients presented with large splenomegaly (spleen palpable ≥10 cm below the left costal margin); the median TSS was 10 (TSS ≥ 20 in 31.7% of the patients). The median duration of the temporary drug discontinuation was 2 months (range, 0.5-71.1 months), and it was slightly shorter in patients who discontinued because of toxicity (median, 1.3 months) in comparison with patients who had an inadequate response (median, 8.2 months; $P = .05$). Although 38 patients (63.3%) rechallenged ruxolitinib within 3 months after the first discontinuation, 12 patients (20%), 3 patients (5%), and 7 patients (11.7%) rechallenged the drug after 3 to 6 months, 6 to 12 months, and more than 12 months, respectively.

Between ruxolitinib discontinuation and rechallenge, 80% of RUX-again patients received no therapy or only palliation (including corticosteroids and/or hydroxyurea and/or recombinant erythropoietin); 11.7% switched to investigational agents (including alternative *JAK2* inhibitors, telomerase inhibitors, and/or antifibrotic agents); and 3.3% and 5% underwent splenectomy and ASCT, respectively. In comparison with the disease status when ruxolitinib was stopped, at rechallenge, there was a significant increase in the number of patients with larger splenomegaly and a higher TSS (Table 2). These variations between stop and rechallenge remained significant even when we considered the following categorical variables: spleen ≥ 10 cm below the costal margin ($P = .01$) and TSS ≥ 20 ($P < .001$). The ruxolitinib dose was lower at restart versus the first stop ($P = .04$); however, the dose reductions were minimal because the variation in patients with a ruxolitinib dose > 10 mg twice daily was not significant ($P = .21$), with 73.3% of patients remaining in the same category at both time points. Also, no dose differences were observed between patients who discontinued because of a lack/loss of spleen response and patients who discontinued because of toxicity ($P = .44$). Four patients were not evaluable for spleen length because they underwent splenectomy before the start of ruxolitinib (n = 2) or before ruxolitinib rechallenge (n = 2).

During the rechallenge period, 44.6% and 48.3% of the patients had spleen and symptom improvements, respectively, and there was a significant increase in the number of patients with a TSS reduction ($P = .01$); 12 patients (20%) continued ruxolitinib with a stable/worsening spleen size but an improvement in the TSS. Conversely, 26.8% and 20% of the patients had increases in spleen size and in symptoms, respectively.

Notably, patients who rechallenged ruxolitinib with a dose > 10 mg twice daily had a higher probability of achieving reductions of spleen length (subdistribution hazard ratio [SHR], 2.19; 95% CI, 0.99-4.86; $P = .05$) and TSS (SHR, 2.67; 95% CI, 1.20-5.93; $P = .02$). Conversely, no association was found between spleen/TSS reductions and an age ≥ 65 years ($P = .81/P = .17$), male sex ($P = .34/P = .84$), a hemoglobin level < 10 g/dL ($P = .70/P = .62$), a platelet count < 100×10^9/L ($P = .34/P = .64$), a spleen ≥ 10 cm below the costal margin ($P = .67/P = .38$), a TSS ≥ 20 ($P = .88/P = .45$), a duration of ruxolitinib discontinuation > 3 months ($P = .20/P = .29$) or > 12 months ($P = .20/P =$

TABLE 2. Clinical and Laboratory Characteristics at the First Stop of Ruxolitinib, at Rechallenge, and at Last Contact on Ruxolitinib

Characteristic	At Discontinuation, Median (Range)	At Rechallenge, Median (Range)	No. of Patients With Increased Values	No. of Patients With Decreased Values	No. of Patients With Stable Values	P^a	At Last Contact on Ruxolitinib, Median (Range)	No. of Patients With Increased Values	No. of Patients With Decreased Values	No. of Patients With Stable Values	P^b
Hemoglobin, g/dL	9.5 (5-15.9)	9.1 (6-16.2)	31	25	4	.22	9.2 (5.3-15.9)	22	32	6	.17
Leukocytes, $\times 10^9$/L	7 (1.7-81.7)	8 (1.8-75)	33	20	7	.19	8.1 (1.3-90)	26	27	7	.63
Platelets, $\times 10^9$/L	93 (8-870)	141 (45-1305)	34	22	4	.01	114 (4-375)	15	43	2	<.001
Spleen length below costal margin, cm	8 (0-28)	10 (0-29)	31	8	17	<.001	8 (0-34)	15	25	16	.12
Total Symptom Score	10 (0-52)	20 (0-66)	34	10	16	<.001	10 (0-85)	12	29	19	.01
Ruxolitinib dose, median, mg bid	10 (2.5-25)	10 (2.5-25)	12	24	24	.04	10 (1.25-20)	18	10	32	.20

Abbreviation: bid, twice daily.
The doses of 2.5 and 1.25 mg bid stand for 5 mg once daily and 5 mg every other day, respectively.
[a]*P* values from Wilcoxon signed-rank tests assessing the variations of variables between the first stop and rechallenge.
[b]*P* values from Wilcoxon signed-rank tests assessing the variations of variables between rechallenge and last contact on ruxolitinib. Four patients were not evaluable for spleen length because they underwent splenectomy.

Figure 2. Overall survival for RUX-again and RUX-stop patients. Overall survival was estimated from the date of the first/only ruxolitinib (RUX) discontinuation to last contact. RUX-again patients were those who rechallenged RUX after a first discontinuation; RUX-stop patients were those who permanently discontinued RUX.

.37), the use of other therapies before rechallenge ($P = .20/P = .37$), or the cause of ruxolitinib discontinuation (resistance vs toxicity; $P = .93/P = .87$).

Overall, 31 patients (51.7%) permanently discontinued ruxolitinib because of death (32.3%), a lack of response (29%), hematological toxicity (19.3%), MF progression (6.5%), infection (3.2%), or bleeding (3.2%); 6.5% of patients discontinued while having a good response to undergo ASCT. The proportions of RUX-again patients who permanently discontinued ruxolitinib were 20%, 33.3%, and 48.3% at 6, 12, and 24 months, respectively. The median follow-up from the permanent discontinuation of ruxolitinib to last contact was 10.9 months (range, 1.2-45 months). Among the 21 living patients at the time of the second discontinuation, most (54.5%) received no or palliative therapy, whereas 3 were treated with an investigational *JAK2* inhibitor, and 2 underwent ASCT.

Outcomes of Patients According to Ruxolitinib Rechallenge

Among the 159 RUX-stop patients, 68.5% received no therapy or only palliation, including corticosteroids and/or hydroxyurea; 15.1% received ASCT, 10.7% received investigational agents, and 5.7% underwent splenectomy. The use of other treatments excluding ruxolitinib was

comparable in RUX-again and RUX-stop patients (investigational agents, log-rank $P = .28$; ASCT, log-rank $P = .09$; splenectomy, log-rank $P = .08$).

From the date of first/last ruxolitinib discontinuation, a total of 25 RUX-again patients (41.7%) and 105 RUX-stop patients (66.0%) died. Causes of death were progressive MF (36.2%), infections (16.2%), second primary malignancies (8.5%), progression to blast phase (6.9%), and other causes (32.2%). Causes of death were comparable in RUX-again and RUX-stop patients ($P = .32$).

The overall survival rate for the total cohort was 68.6% and 40.6% at 1 and 3 years, respectively. Notably, RUX-again patients had significantly longer survival than RUX-stop patients with median survival times of 41.1 and 23.7 months, respectively, in the 2 cohorts (log-rank $P = .004$; Fig. 2). However, overall survival was comparable in patients who discontinued ruxolitinib because of a lack/loss of response and patients who discontinued ruxolitinib because of toxicity (median survival, 27.9 and 27.6 months, respectively; $P = .63$). When we compared patients with a lack or loss of response, no survival difference was observed ($P = .16$).

DISCUSSION

According to the 2020 National Comprehensive Cancer Network guidelines, possible medical alternatives beyond palliation, in cases of resistance or intolerance to ruxolitinib, include the use of fedratinib and investigational agents.[25]

This real-world study was completed before the availability of fedratinib in Europe, and it shows that ruxolitinib rechallenge was quite common after initial ruxolitinib failure, involving almost 30% of patients who discontinued the drug in the chronic phase. In the absence of alternative *JAK2* inhibitors in routine practice, rechallenge, representing an easily viable option, particularly in intolerant patients, was attempted early and before other therapeutic approaches in most cases.

The temporary discontinuation generally caused a significant increase in the disease burden, which reflected a loss of residual control activity not only in intolerant patients but also in resistant patients. Analogously, a ruxolitinib rebound syndrome (RDS), attributed to an acute rebound of cytokine storm soon after ruxolitinib discontinuation, has been documented in many resistant patients.[26-30] In this regard, we previously observed that thrombocytopenia (platelet count $< 100 \times 10^9$/L) and a large spleen (palpable ≥10 cm below the costal margin) at the stopping of ruxolitinib were significantly associated with a higher probability of RDS. The association between a higher disease burden and RDS was interpreted as a nonnegligible activity of *JAK2* inhibition in at least some patients with refractory MF, and this may explain the observed efficacy of rechallenge also in refractory patients.[26]

After the rechallenge, clinical responses were achieved by almost 50% of the patients. This therapeutic efficacy is deemed to be based on a resensitization to *JAK2* inhibition through different mechanisms, including restoration of homodimeric JAK-STAT signaling.[31] However, other pathways may contribute to resistance to ruxolitinib and may be overcome with drug discontinuation.[32] The finding that only the use of high ruxolitinib doses (>10 mg twice daily) was associated with an increased probability of spleen and symptom improvements corroborates the positive relationship between dose and response shown in the COMFORT studies and by real-world evidence.[2,33,34]

Notably, the lack of an association between rechallenge efficacy and reasons for discontinuation probably stems from the fact that intolerance and resistance often overlap (ie, the patient does not achieve or loses a response because ruxolitinib is administered at suboptimal doses or intermittently on account of intolerance). In other cases, intolerance reveals a more aggressive disease and/or an intrinsic frailty of the patient (ie, concomitant comorbidities, polypharmacy, more frequent infections, and greater thrombotic-hemorrhagic risk), and this results in reduced survival. In the absence of alternative treatment strategies, ruxolitinib rechallenge may, therefore, be considered in virtually all patients with active disease. However, the durability of ruxolitinib rechallenge was quite short, with approximately 50% of RUX-again patients discontinuing the drug permanently at 2 years. This observation may suggest strict clinical follow-up of patients during the rechallenge and the rapid implementation of alternative therapeutic strategies when required.

Finally, survival seemed to be improved in RUX-again patients compared with RUX-stop patients despite the use of investigational agents, and ASCT was comparable in the 2 groups. This finding complements the survival benefit results observed in the COMFORT studies and extends real-world evidence of a survival advantage for patients treated with novel agents, including ruxolitinib rechallenge, after ruxolitinib failure.[3,4]

The main constraint of this study is its retrospective nature. Indeed, suboptimal management or dosing of ruxolitinib, inadequate recognition of failure or intolerance of ruxolitinib, and poor assessment of drug compliance

cannot be ruled out and may have contributed to premature drug discontinuations in real life. Also, it was not possible to ascertain in which patients the rechallenge was intentional and in which patients it occurred in the absence of a premeditated therapeutic strategy, mostly in reaction to toxicity. We observed that the cause of ruxolitinib discontinuation (resistance vs toxicity) as well as its duration (longer than 3 or 12 months) and the use of other therapies did not affect the efficacy of ruxolitinib rechallenge. However, the distinction between these 2 clinical situations may be relevant because the success rate of an intentional ruxolitinib holiday might be higher.

Nonetheless, the substantial number of included patients, the cooperation of hematology centers with a particular focus on MF, and the accurate revision of each case history may partially compensate for these intrinsic shortcomings. We acknowledge that this limitation can hardly be avoided when one is dealing with a rare condition such as MF and a specific subpopulation such as ruxolitinib-treated patients. On the other hand, after the approval of ruxolitinib for MF therapy, retrospective studies may represent the only and valuable source of comprehensive data and lead to personalized therapy.

Overall, these findings provide important real-life evidence that ruxolitinib rechallenge may be effective after initial ruxolitinib failure, with clinical improvements achieved in a not negligible portion of patients. However, ruxolitinib rechallenge was used mainly in intolerant patients and was associated with a high rate of permanent drug discontinuation. The survival advantage observed in RUX-again patients highlights the role of appropriate treatment strategies and ruxolitinib use in outcomes. Other *JAK2* inhibitors and alternative drugs are currently under clinical investigation and may soon broaden the therapeutic scenario of MF further.[35] Future real-world evidence will possibly clarify whether the use of ruxolitinib rechallenge will be reduced or abandoned with the advent of new treatments in clinical practice and what criteria should be used to select a patient for one treatment or another.

FUNDING SUPPORT

This study was supported by AIL Bologna.

CONFLICT OF INTEREST DISCLOSURES

Francesca Palandri, Mario Tiribelli, and Malgorzata M. Trawinska report consultancy for and honoraria from Novartis; Tiribelli also reports personal fees from Bristol-Myers Squibb/Celgene, Pfizer, and Incyte. Giulia Benevolo reports honoraria from Novartis, Janssen, and Amgen. Alessandra Iurlo, Massimo Breccia, Elisabetta Abruzzese, and Massimiliano Bonifacio report honoraria from Novartis, Bristol-Myers Squibb, Pfizer, and Incyte; Bonifacio also reports personal fees from Amgen, and Abruzzese also reports other from Takeda. Gianpietro Semenzato reports honoraria from AbbVie, Roche, and Takeda. Francesco Cavazzini reports honoraria from Novartis, Incyte, and Pfizer. Roberto Latagliata reports honoraria from Novartis, Celgene, Bristol-Myers Squibb, and Janssen. Roberto M. Lemoli reports honoraria from Gilead, Novartis, Sanofi, and Miltenyi; personal fees from AbbVie, Janssen, Jazz, Daiichi Sankyo, and Servier; and research funding from Celgene. Florian H. Heidel reports consultancy for and research funding from Novartis, CTI, and Celgene. Nicola Polverelli reports personal fees from Novartis. Michele Cavo reports consultancy for and honoraria from Janssen, Bristol-Myers Squibb, Celgene, Sanofi, GlaxoSmithKline, Takeda, Amgen, Oncopeptides, AbbVie, Karyopharm, and Adaptive. Giuseppe A. Palumbo reports honoraria from Novartis, Celgene, Janssen, Amgen, Hospira, and Teva. Elena Masselli reports personal fees from Uvet GB SPA, Iqvia Solutions SRL, and Proeventi SRL. The other authors made no disclosures.

AUTHOR CONTRIBUTIONS

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