VIA MEDICA
REVIEW ARTICLE

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Ruxolitinib in the treatment of patients with myelofibrosis — questions and answers

This is a translation of an article published in Polish in the journal „Hematologia" 2018, vol. 9, no. 4, 269–284. DOI: 10.5603/Hem.2018.0035

Introduction

Myelofibrosis (MF) is a clonal disease, arising as a result of somatic mutations in pluripotent stem cells. This leads to proliferation of atypical megakaryocytes and disfunction of the bone marrow microenvironment. Deregulation of JAK-STAT (Janus kinase — signal transducers and activators of transcription) pathway plays a key role in MF pathogenesis. Most patients carry mutation of the tyrosine kinase gene *JAK2* V617F in exon 14. In patients with wild-type *JAK2* gene, about 10% have mutation in the *MPL* W515L/K gene coding receptor for thrombopoietin, and in 80% of the remaining patients a mutation in the calreticulin gene (*CALR*) can be detected. All three described mutations lead to constitutive activation of JAK-STAT pathway, which results in increased secretion of proinflammatory cytokines, including interleukin 8, 10, 15, and tumour necrosis factor alpha (TNFα), as well as increased secretion of growth factors: vascular endothelial growth factor (VEGF), basic fibroblast growth factor (bFGF), platelet-derived growth factor (PDGF), and transforming growth factor beta (TGFβ). Excess of enumerated particles increases fibrosis, induces extra-medullary haematopoiesis, stimulates angiogenesis, and raises constitutional catabolism. Lack of either of these three mutations, found in about 10% of patients, is correlated with poor prognosis. Besides the presence of the described "driver" mutations, several types of mutations in genes regulating epigenetic changes can be found (including *ASXL1, EZH2, TET2, DNMT3A, IDH1/2, SRFS2, SRF3B1, TP53*). Detection of at least one mutation in *ASXL1, EZH2, SRSF2*, and *IDH1/2* genes determines high molecular risk (HMR), associated with shorter overall survival (OS) and higher risk of blastic transformation.

Described clinical and molecular features were incorporated in the newest prognostic scales, which supports optimal clinical management of patients with MF. In 2009, the International Working Group — Myeloproliferative Neoplasms Research and Treatment (IWG-MRT) collaboration developed the International Prognostic Scoring System (IPSS) scale, based on five independent progression risk factors assessed at the time of MF diagnosis. This included: age over 65 years; presence of systematic symptoms; haemoglobin (Hb) concentration lower than 10 g/dl; hyperleukocytosis over 25 G/l; and the presence of at least 1% of blasts in peripheral blood smear. The IPSS scale was subsequently expanded into Dynamic IPSS (DIPSS), which included the possibility of acquisition of the aforementioned risk factors during the course of the disease, and provides prognostic stratification at any point of MF duration. In the DIPSS Plus scale, three additional independent prognostic factors were included: dependency on blood transfusions; unfavourable karyotype (trisomy 8;

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Translation: lek. Maciej Kawecki
Oncology in Clinical Practice, 2019, Vol. 15, No. 3, 167–179, DOI: 10.5603/OCP.2019.0021, Copyright © 2019 Via Medica, ISSN 2450–1654

monosomy 7/7q–; i(17q); inv(3); monosomy 5/5q– or 12p–; rearrangement of 11q23); and thrombocytopaenia (platelet count lower than 100 G/l).

Until recently, there was no drug to slow MF progression or to control systemic symptoms. Ruxolitinib — an inhibitor of JAK1/JAK2 kinase — is the first and, at present, only registered drug for MF that has changed this calamitous situation. It was approved by the Food and Drug Administration (FDA) in the USA in 2011 to treat patients with intermediate- or high-risk according to IPSS. In 2012, the European Medicines Agency (EMA) registered ruxolitinib in the EU to treat patients with MF, who had splenomegaly and/or systemic symptoms. Both decisions were based on the results of two phase III trials: COMFORT-I and COMFORT-II. The trials proved effectiveness of ruxolitinib in reducing splenic volume and in decreasing constitutive symptoms in MF patients with and without V617F mutation. Combined analysis of OS after three years of follow-up showed over 30% reduction in death risk in patients receiving ruxolitinib when compared to best available therapy or placebo. The described results and further statistical analyses led to the reimbursing ruxolitinib in Poland on 1st January 2017. Now the drug is available as a part of the Polish National Health Fund Drug Program, which includes patients with both primary and secondary MF, intermediate (2) or high IPSS risk, splenomegaly (spleen palpable ≥ 5 cm under ribs and/or splenomegaly present in ultrasound examination), and systemic symptoms.

Of utmost importance, ruxolitinib can be used in MF patients scheduled to receive allogenic haematopoietic stem cell transplantation (allo-HSCT). A decrease in concentration of proinflammatory cytokines, reduction of systematic symptom burden, shrinkage of spleen, and improvement of physical performance achieved before transplantation can lead to lower mortality and better outcomes associated with bone marrow transplant. According to European Leukaemia Net (ELN) and European Society for Blood and Marrow Transplantation (EBMT) guidelines, treatment with ruxolitinib should be initiated at least two months before a planned transplantation. The ruxolitinib dose should be gradually reduced 5–7 days before conditioning and withdrawn one day prior to the procedure. Retrospective analyses suggest that the presence of HMR mutations significantly reduce duration of response to ruxolitinib. Therefore, in patients with HMR mutations, who are qualified for bone marrow transplant, treatment with ruxolitinib should be restrained to the period before transplantation, without postponement of this potentially curative procedure.

However, ruxolitinib can lead to numerous adverse events, both haematological and non-haematological. Knowledge of the toxicity profile and proper adverse event management is required for effective and safe treatment. The article below presents the clinical aspects of ruxolitinib treatment in patients with MF, including groups with different clinical, laboratory, and pathological features. Expert opinions are supported with literature data and provide valuable advice for haematologists in their daily practice.

Ruxolitinib in patients with liver injury

The mean age of patients with MF is 65.9 years and with *polycythaemia vera* (PV) — 60.8 years [1]. This population is characterised by numerous comorbidities, including the presence of liver injury detected in physical examination, laboratory results, or in radiological imaging. With rising age, the rate of patients with hepatopathy increases, mostly due to toxic (alcohol, drugs) or metabolic (diabetes, hyperlipidaemias) factors. A significant proportion of hepatopathies arise from common infections with hepatitis B virus (HBV) or hepatitis C virus (HVC). Another significant factor responsible for hepatopathy in patients with myeloproliferative diseases is extra-medullar haematopoiesis, usually in the liver. As a result, hepatomegaly might be present in more than half of all patients with MF. One of the most common non-haematological adverse events observed with ruxolitinib in registration trials was an increase in aminotransferases activity. This might be observed in about 20–30% of treated patients. Additionally, ruxolitinib elimination might be prolonged in patients with liver insufficiency [2].

Evaluation of liver function is required before ruxolitinib treatment initiation. Laboratory studies should include aminotransferase activity and bilirubin concentration. Patients qualified for ruxolitinib treatment should have bilirubin concentration not higher than two-fold of the upper limit of normal (ULN) and alanine and aspartate aminotransferase activity lower than 2.5-fold of ULN. In patients with aminotransferases and/or bilirubin elevated below mentioned thresholds, detailed diagnostics should be undertaken. This is crucial because ruxolitinib treatment might lead to further increases in aminotransferase activity due to its hepatotoxic potential. Patients with elevated liver exams should be evaluated for the presence of active HBV or HCV hepatitis (HBsAg, anti-HBc, anti-HCV). Positive results should mitigate quantitative assessment for HBV-DNA and HCV-RNA. Infectious diseases specialist consultation might be required. Another possible cause of liver injury might be abuse of non-steroidal anti-inflammatory drugs. The most important task should be withdrawal of the over-used drugs. Liver regeneration may be supported with phospholipids (Esseliv forte, Essentiale forte, Essentialne Vital) or silymarin preparations (Sylimarol Vita). In patients with primary bone marrow fibrosis, who require numerous blood transfusions, secondary

haemochromatosis might be considered as a source of liver damage. Such patients should be monitored for ferritin concentration.

Patients with significant liver injury, defined as an increase in aminotransferase ≥ 2.5-fold ULN and increase in bilirubin concentration ≥ 2-fold ULN, require 50% reduction of ruxolitinib dose. The most important factor influencing initial ruxolitinib dose is the number of platelets (PLT). For example, for a patient with alanine aminotransferase increase of 1.5-fold over ULN and PLT number of 250 G/l, the initial treatment dose should be 10 mg of ruxolitinib administered twice daily. Monitoring with complete blood count, aminotransferase activity, and bilirubin concentration is required every 1–2 weeks for the first six weeks of treatment. If increased aminotransferase activity persist or if the PLT number decreases, the ruxolitinib dose should be again reduced.

Interactions of ruxolitinib with other drugs

Cytochrome P450 (CYP) inhibitors

Studies evaluating ruxolitinib *in vivo* showed that CYP3A4 is the main isoenzyme responsible for its metabolism. Patients treated with ruxolitinib may receive other drugs metabolised through the same enzymatic pathway. If ruxolitinib is administered simultaneously with strong CYP3A4 inhibitors or double CYP2C9 and CYP3A4 inhibitors (e.g. fluconazole), ruxolitinib dose should be reduced by 50% in two daily doses. Intensified monitoring of haematological parameters and regular physical examination screening for liver injury is advised. Strong CYP3A4 inhibitors include: boceprevir, clarithromycin, indinavir, itraconazole, ketoconazole, lopinavir, ritonavir, mibefradil, nefazodone, nelfinavir, posaconazole, saquinavir, telaprevir, telithromycin, and voriconazole.

The ruxolitinib dose should not be reduced if the drug is given simultaneously with weak or moderate CYP3A4 inhibitors. These include ciprofloxacin, erythromycin, amprenavir, atazanavir, diltiazem, and cimetidine. However, patients should be closely monitored for potential cytopaenia when moderate CYP3A4 inhibitor treatment is initiated. Concomitant treatment with ruxolitinib and cytoreductive drugs or haematopoietic growth factors was not studied, and therefore the safety and efficiency of such treatment is unknown. Selective serotonin reuptake inhibitors (SSRI) increase serum ruxolitinib concentration because they inhibit activity of CYP3A4 isoenzymes. Such SSRIs include: fluoxetine, fluvoxamine, sertraline, and paroxetine. In patients receiving ruxolitinib, antidepressants with a mode of action different from SSRI are advised.

CYP3A4 inductors

Patients requiring chronic treatment with CYP3A4 inductors (such as avasimibe, carbamazepine, phenobarbital, phenytoin, rifabutin, rifampicin) should be closely monitored. Changes in CYP3A4 activity have limited impact on pharmacodynamics of ruxolitinib and is insignificant from a clinical standpoint. The dose of ruxolitinib can be gradually increased, considering the safety and effectiveness of treatment.

Infections

In retrospective analyses of patients with MF treated with ruxolitinib, about 20% develop infections, 90% of which are bacterial. Factors associated with infections are age over 65 years and concomitant treatment with corticosteroids.

Increased risk of infections with atypical strains of mycobacteria, pneumocystis, and reactivation of type B hepatitis should be noticed. Screening for human immunodeficiency virus (HIV), HBV, and HCV before ruxolitinib initiation is strongly encouraged. All patients with MF treated with ruxolitinib in clinical trials were offered annual influenza vaccination and pneumococcal vaccination because ruxolitinib treatment may result in immunodeficiency due to its potential to impair functioning of T cells, dendritic cells, and natural killer (NK) cells. For the same reason, patients treated with ruxolitinib must not receive live vaccinations. Fungal infections should be closely controlled because most antifungal drugs are CYP2C9 and CYP3A4 isoenzyme inhibitors and can lower therapeutic activity of ruxolitinib. Despite no pharmacological interaction between ruxolitinib and steroids, their concomitant usage is not recommended due to the unfavourable impact on cell-mediated immunity. Opportunistic infections, such as mycobacterial and pneumocystis infections, were described in patients treated with ruxolitinib and steroids. Similarly, despite no interaction between ruxolitinib and thalidomide described, both drugs have myelosuppressive potential and therefore patients receiving them simultaneously should be carefully monitored.

Ruxolitinib treatment in patients with anaemia. When to reduce the dose and when to withdraw therapy?

Anaemia is present in 35–54% of patients with MF at diagnosis and is considered an unfavourable prognostic factor [3]. With the course of the disease, the rate of anaemia rises and after a year is present in 47–64% of patients [3–6]. Ruxolitinib's mode of action, as well as the pathophysiological mechanism present in MF, result

in anaemia (with Hb concentration lower than 10 g/dl), being one of the most common adverse events. In both the COMFORT-I and COMFORT-II trials, patients receiving ruxolitinib experienced a decrease in Hb concentration during the first 12 weeks, with a nadir between the 8th and 12th week. Additionally, in both trials after 24 weeks of treatment, the Hb concentration increased to over 10 g/dl and stabilised at that level, which was independent of blood transfusions or ruxolitinib dose reductions [6–9]. Long-term observation from the COMFORT-I trial suggests that the incidence of new anaemia episodes grade 3 or 4 according to Common Terminology Criteria For Adverse Events (CTCAE) decreased with the length of treatment [10] and is not significantly higher than in patients receiving placebo [11]. These observations are confirmed by routine clinical practice. Anaemia is present in 70–75% of patients treated with ruxolitinib, usually during the first three months of treatment [12]. In most patients, the Hb concentration rises and stabilises thereafter. Analyses of data obtained in the COMFORT trials indicate that ruxolitinib-induced anaemia is not a negative prognostic factor and does not affect OS. For most experts who treat patients with MF, an Hb concentration decrease in the first weeks of ruxolitinib treatment is not an indication to reduce the dose or withdraw ruxolitinib, because this may lead to recurrence of symptoms present at the treatment initiation, usually within the first 10 days [12]. In patients without anaemia at the time of treatment initiation (e.g. with Hb concentration of 12 g/dl), who experienced decrease of Hb concentration to about 8.5 g/dl along with benefit from ruxolitinib, continuation of treatment with a possible dose reduction can be recommended. Nevertheless, most experts stress that the degree of anaemia is rarely the only reason for dose adjustment. In patients with anaemia and Hb concentration lower than 10 g/dl at the time of therapy initiation, the starting dose should be 10 mg twice daily. In patients dependent on blood transfusions, the recommended starting dose is 5 mg twice daily with a possible increase if tolerated. The initial three months of treatment are usually crucial to adjust doses for each patient [12].

A subgroup of patients do not achieve stabilisation of anaemia after the first three months of ruxolitinib treatment. Most experts agree that ruxolitinib dose reduction due to anaemia or blood transfusion dependency is not necessary, unless the decrease in Hb is substantial (e.g. from 11 g/dl to 6 g/dl) [12]. A mild decrease in Hb concentration (e.g. from 11 g/dl to 9 g/dl) is usually acceptable if the patient does not develop significant fatigue. In the case of Hb decrease from, as an example, 10 g/dl to less than 8 g/dl and concomitant significant fatigue, the decision about dose reduction should be preceded by consideration of whether symptoms associated with anaemia provide more burden than symptoms related to MF. The decision about dose reduction might be influenced by the patient's age. The treatment in younger patients might be more intensive than in patients older than 70 years, who require a more cautious approach. In patients who have low Hb concentration despite blood transfusions, along with a significant fatigue, and who prefer dose reduction despite adequate PLT number, dose reduction from the initial 20 mg twice daily to 15 mg or even 10 mg twice daily might be considered. The reduced dose should be continued with a close follow-up as long as the patient maintains response [12]. If an increase in Hb is observed, ruxolitinib dose escalation should be considered. If no change in Hb concentration is seen, erythropoiesis-stimulating agents (ESA) might be considered. In patients who develop rapid and significant decrease of Hb (to less than 6 g/dl) after prolonged treatment (e.g. 6–8 months) or who require blood transfusion more often than biweekly and who have recurrence of systemic symptoms and limited reduction of spleen volume, ruxolitinib withdrawal may be considered. The decision regarding dose reduction or ruxolitinib withdrawal should be taken individually, after discussion with the patient. Some patients might prefer continuation of treatment because it provides substantial reduction of MF symptoms, while others might prefer discontinuation to avoid frequent blood transfusions [12].

Erythropoietin in patients with myelofibrosis treated with ruxolitinib

Erythropoiesis-stimulating agents acts through the same pathway as endogenous erythropoietin, the concentration of which increases in patients treated with ruxolitinib as a result of *JAK2* pathway inhibition and suppression of proliferation and final differentiation of erythropoietic precursor cells. Therefore, it might be expected that ESA administration would provide limited benefit. However, it seems that for the increase in mean number of circulating erythrocytes the erythropoietin serum concentration is less important than its mean serum half-life time. Most currently used ESAs are characterised by a prolonged half-life when compared to endogenous erythropoietin and therefore may offer clinical benefit. ESA were used in 13 from 146 patients (9%) treated with ruxolitinib in COMFORT-II trial. Darbepoetin alpha was administered to three patients in doses 40–300 μg, 150–300 μg, and 500 μg; epoetin alpha was used in nine patients in doses between 10 and 40 thousand units; another erythropoietin preparation was administered to one patient at doses between 10 and 20 thousand units. Mean doses of ruxolitinib administered to patients receiving ESA and not receiving ESA were similar. Additionally, rates of patients requiring dose reductions were also similar. Due to the limited

number of this population, no statistical analyses comparing patients receiving and not receiving ESA were possible. Compared with lowest Hb concentration before ESA initiation, the lowest Hb concentration during first three months of ESA administration was increased in three patients, stable in seven patients, and lower in two patients. After three months of ESA treatment, Hb increase was observed in six patients (mean rise 7 g/dl) and Hb decrease in two patients (no data was reported regarding another five patients). In the analogic period, mean blood transfusion number decreased in two patients, was stable in one patient, and increased in three patients. Seven patients, who were independent of blood transfusion before ESA initiation, remained independent after three months of ESA treatment. Six weeks before ESA initiation grade 3 and 4 anaemia (according to CTCAE) was noticed in 10 among all 13 patients (77%). After six weeks of ESA treatment, in seven out of 13 patients (54%) anaemia grade decreased to grade 2 according to CTCAE. Among serious adverse events reported in eight patients receiving ESA, one episode of pulmonary embolism was judged to be ESA-related [3]. Results of other clinical trials indicate that ESA administration has limited effectiveness in MF patients who are blood transfusion dependent, have significant splenomegaly, have endogenous erythropoietin concentration of over 125 units/l, or have homozygotic mutation of *JAK2* gene [9]. No patient with normal endogenous erythropoietin concentration responded to ESA in another trial [11]. In a different trial undertaken in Mayo Clinic, no difference in response to ESA was seen regardless of initial erythropoietin concentration and was generally considered to be low (in 23% patients) [11]. Doubts regarding safety and possible association with leukaemic transformation have led to ESA being unrecommended in patients with MF, who are blood transfusion dependent or who have Hb concentration higher than 10 g/dl before treatment initiation [9, 11, 13]. The benefit seen in some patients receiving ESA in the COMFORT-II trial might be due to the prolonged ESA half-life compared to endogenous erythropoietin, and to the relatively short half-life of ruxolitinib. Obtained results suggest that in this group of patients ESA can be administered safely, without any negative impact on ruxolitinib effectiveness, and might be used to maintain ruxolitinib-related anaemia in the future.

Ruxolitinib in patients dependent on blood transfusions and with low PLT count

Anaemia and thrombocytopaenia are clinical manifestations of the advanced, fibrotic phase of myelofibrosis. The main mechanism leading to the development of these symptoms is suppression of erythropoietic and thrombopoietic precursors by progressive fibrosis in bone marrow and excessive degradation of erythrocytes and thrombocytes in an enlarged spleen. PLT count lower than 100 G/l, Hg concentration lower than 10 G/l, and blood transfusion dependency are poor prognostic factors and were included in IPSS, DIPSS, and DIPSS Plus classifications [14]. It is estimated that at the time of MF diagnosis anaemia with Hb concentration lower than 10 g/dl is present in 35–50% of patients and thrombocytopaenia with PLT count lower than 100 G/l is present in about 25% of patients [15]. Patients who begin treatment with ruxolitinib usually experience anaemia and thrombocytopaenia as a result of inhibition of JAK2 kinase-dependent erythropoiesis and thrombopoiesis. Both anaemia and thrombocytopaenia are strictly correlated with ruxolitinib dose. Patients with a tendency to develop thrombocytopaenia and anaemia should be carefully monitored. Avoiding significant decrease of platelet or erythrocyte count may limit the risk of serious adverse events, especially haemorrhages. Dose reduction is the most appropriate way of action in case of significant anaemia and/or thrombocytopaenia. Even temporary ruxolitinib withdrawal should be avoided because this may result in a flair-effect. Patients with severe decrease in Hb concentration and/or PLT count should receive packed red blood cells and/or platelet concentrate.

The first data regarding frequency of anaemia and thrombocytopaenia in patients treated with ruxolitinib came from the COMFORT-I and COMFORT-II trials. This evidence was the basis for recommendations regarding ruxolitinib dose reductions and interruptions. Because the first 8–12 weeks of treatment are associated with the highest risk of thrombocytopaenia, the initial ruxolitinib dose should be based on pre-treatment PLT count (PLT > 200 G/l — 2 × 20 mg, PLT 100 G/l to 200 G/l — 2 × 15 mg, PLT 50 G/l to 100 G/l — 2 × 5 mg). If the PLT count decreases below 50 G/l during ruxolitinib treatment, the dose should be slowly reduced and then, if necessary, ruxolitinib may be withdrawn [16–19]. Subsequent clinical trials (JUMP, EXPAND), which recruited patients with PLT count lower than in COMFORT trials, allowed the development of guidelines for ruxolitinib dose reductions in cases of more sever thrombocytopaenia. In American practice, ruxolitinib is withdrawn after the PLT count falls below 25 G/l, according to the Summary of Product Characteristics accepted by FDA. Because dose reduction might have a negative impact on treatment effectiveness, the highest tolerable dose should be reintroduced once the grade of toxicity allows [16].

The COMFORT trials showed that 61% of patients receiving ruxolitinib, who had normal pre-treatment haemoglobin concentration, developed anaemia, and

69% of patients with pre-treatment experienced anaemia worsening. Red blood cell parameters achieve their lowest point usually between eight and 12 weeks after treatment initiation and return to baseline after 24 weeks of therapy. In the case of anaemia, even with very low Hb concentration, ruxolitinib withdrawal is not recommended because the anaemia can be managed with blood transfusions and dose reductions, although this was not recommended in COMFORT trials. Exploratory analysis of the COMFORT trial data showed that, despite the fact that any degree of pre-treatment anaemia is a negative prognostic factor, anaemia associated with ruxolitinib treatment does not affect the patient's prognosis [15].

Maintenance of optimal ruxolitinib dose, adjusted to PLT count and Hb concentration, requires regular evaluation of complete blood count (CBC), especially during expected PLT and Hb nadir (between eight and 12 weeks after therapy initiation). Bi-weekly laboratory assessment can be recommended in all patients, even often in patients with low PLT count, who are dependent on blood transfusions. Adequate, regular laboratory evaluation and skilful dose maintenance might be crucial for successful ruxolitinib treatment [15].

IPSS and DIPSS — practice *versus* Drug Program. Which scale to use and how often to evaluate?

In 2009 the IWG-MRT group analysed a cohort of 1054 patients with newly developed MF and developed the IPSS scale. The analysis discriminated five independent progression risk factors: age over 65 years, presence of systemic symptoms, Hb concentration lower than 10 g/dl, hyperleukocytosis over 25 G/l, and the presence of at least 1% of blasts in a leukogram. Every factor was attributed one point. The number of points classifies patients to a group with low (0 points), intermediate-1 (1 point), intermediate-2 (2 points), or high risk (≥ 3 points), with median OS of, respectively, 135, 95, 48, and 27 months [20].

Expansion of IPSS, which was developed for patients before treatment initiation, led to the DIPSS scale, which incorporated acquisition of risk factors during the course of disease and can be used at any time. The DIPSS scale included the same parameters as the IPSS scale, with a 2-point value attributed to anaemia. The number of points classifies patients to groups with low (0 points), intermediate-1 (1–2 points), intermediate-2 (3–4 points), and high (5–6 points) risk, with median OS of, respectively: not reached, 168, 48, and 18 months [21]. The DIPSS scale can also assess risk of transformation to acute myeloid leukaemia: it can be estimated at, respectively, 0.3, 0.7, 2.6, and 8.6 cases per 100 patient-years [22].

In the newer DIPSS Plus scale an additional three independent prognostic factors were included: blood transfusions dependency, unfavourable karyotype (trisomy 8; monosomy 7/7q–; i(17q); inv(3); monosomy 5/5q– or 12p–; rearrangement of 11q23), and thrombocytopaenia (PLT count lower than 100 G/l) [23].

The Polish National Health Fund Drug Program requires attribution of potential patients to intermediate-2 or high-risk groups in the IPSS scale (which is based on the results of registration trials). If the patient was previously surveilled and attributed to the low-risk group, reassessment with the IPSS scale is discordant with its basic assumption of evaluation at the time of diagnosis. Patient surveillance should be undertaken with dynamic scales, such as DIPSS and DIPSS Plus. Unfortunately, during administrative controls the Drug Program is interpreted literally (not on the basis of merit), and therefore assessment with a scale other than IPSS during patient qualification can result in a financial fine for a controlled site. Prognostic scales should be actualised during each visit because any sign of progression might require treatment initiation or change. The Drug Program does not require further surveillance with the IPSS scale during treatment.

On a side note, it is worth mentioning that patients with MF, who are potential candidates for allo-HSCT, should not only be assessed with the aforementioned prognostic scales, but also undergo karyotype and molecular risk factor evaluation (including *CALR, JAK2, MPL,* and *ASXL1*). Patients with unfavourable karyotype and/or so-called "triple-negative" patients (without mutation in either *JAK2, CALR*, or *MPL*) with *ASXL1* mutation should be considered as candidates for allo-HSCT even with intermediate-1 risk prognosis in the DIPSS scale.

What, if any, antimicrobial prophylaxis should be administered during ruxolitinib treatment?

Treatment with ruxolitinib may result in immunosuppression, increasing the risk of infectious complications. The pathophysiological nature of this effect is complicated because ruxolitinib results in lower leukocyte count, including granulocytes, with concomitant impairment of lymphocyte T, dendritic cell, and NK cell functioning [24].

Grade 3 and 4 neutropaenia (according to CTCAE) was noted in 7.1% of patients treated with ruxolitinib and in 2% of patients treated with placebo in the COMFORT-I trial [10]. In the COMFORT-II trial, after five-year follow-up, grade 3 and 4 neutropaenia and leucopaenia was noted in 8.9% and 6.3% of patients, respectively (Tab. 1) [25].

Lussana et al. [26] review five phase III randomised clinical trials, six phase IV trials, and 28 case reports and

Table 1. Neutropaenia in patients included in the COMFORT trials (source [25])

	COMFORT-I		COMFORT-II	
	Ruxolitinib (n = 155)	Placebo (n = 151)	Ruxolitinib (n = 146)	BAT (n = 73)
Neutropaenia	19	4	NR	NR
Neutropaenia ≥ grade 3	7	2	NR	NR

BAT — best available therapy; NR — not reported

Table 2. Guidelines for antiviral prophylaxis during ruxolitinib therapy

Pathogen	Laboratory evaluation recommended before treatment initiation	Prophylaxis	Comments
CMV	IgG+, IgM–	Prophylaxis not recommended	CMV-PCR might be considered
EBV	IgG+, IgM–	Prophylaxis not recommended	EBV-PCR might be considered
VZV	IgG+, IgM–	Prophylaxis: acyclovir 2 × 400 mg/d.	
HSV	IgG+, IgM–	Prophylaxis: acyclovir 2 × 400 mg/d.	
HBV	HBsAg+	Prophylaxis: lamivudine 100 mg/d.	
HCV	HCV–, IgG+	No prophylaxis available	

CMV — cytomegalovirus; PCR — polymerase chain reaction; EBV — Epstein-Barr virus; VZV — varicella-zoster virus; HSV — herpes simplex virus; HBV — hepatitis B virus; HCV — hepatitis C virus

showed a statistically significant increase in the risk of shingles in patients treated with ruxolitinib. Data from the COMFORT-I and COMFORT-II trials showed that urinal tract infections grade 3 and 4 developed in 1% of patients, shingles in 4% of patients, tuberculosis in 1% of patients, and sepsis in 3% of patients. Combined analysis of clinical trials demonstrated that the most common infections were: shingles (8%), bronchitis (6%), and urinary tract infections (6%). The most common case reports described tuberculosis (n = 10), HBV reactivation (n = 5), and *Pneumocystis jirovecii* infections (n = 2). Less common cases of bilateral retinitis caused by *Toxoplasma gondii* and confirmed viral leukoencephalopathy were also reported [26].

Available data suggest that the increased infection risk during ruxolitinib treatment can have a clinically significant impact, but no recommended prophylaxis guidelines exist. A limited number of authors formulated practical tips that can be incorporated into clinical practice. Heine et al. [27] proposed undertaking laboratory evaluation aimed at detection of infectious agents before and during ruxolitinib treatment (Tab. 2).

Antibacterial prophylaxis is generally not recommended. Patients with tendencies towards urinary tract infections or bronchopneumonia with granulocyte count lower than 1 G/l may benefit from ciprofloxacin 500 mg administered twice daily until resolution of granulopaenia. Patients with positive results of *Quantiferon* test for *Mycobacterium tuberculosis* are advised to receive isoniazid 300mg daily. No prophylaxis for *Pneumocystis jirovecii* is recommended.

Because ruxolitinib treatment is not associated with an increased risk of fungal infection, no antifungal prophylaxis is recommended. It should be noted that many antifungal drugs are CYP enzyme inhibitors, and their administration might require ruxolitinib dose adjustment.

If ruxolitinib is used concomitantly with strong CYP-3A4 inhibitors or double CYP2C9 and CYP3A4 inhibitors, such as fluconazole, the ruxolitinib dose should be reduced by 50%. A fluconazole dose of 200 mg per day should not be exceeded. If simultaneous administration of ruxolitinib and CYP enzymes inhibitors is required, complete blood count should be evaluated more often — even 1–2 times per week.

Strong CYP3A4 inhibitors, such as clarithromycin, itraconazole, ketoconazole, posaconazole, and voriconazole, also require a 50% reduction of the ruxolitinib dose. Mild and moderate CYP3A4 inhibitors, such as ciprofloxacin and erythromycin, do not require ruxolitinib dose modification, but close monitoring for cytopaenia should be undertaken.

Viral infections, significantly more common in patients receiving ruxolitinib, are a separate issue.

In some cases, antiviral prophylaxis with acyclovir might be considered (Tab. 2) [27].

Can the molecular profile of patient with myelofibrosis affect ruxolitinib effectiveness?

In both COMFORT trials, similar ruxolitinib effectiveness in reduction of splenic volume and control of systemic symptoms was seen in patients with and without V617F mutation [7, 28]. Additional analysis confirmed effectiveness of ruxolitinib in patients with *CALR* mutation [29]. Similar activity of ruxolitinib in both patient groups confirms that the main pathogenetic mechanism behind MF is overactivation of JAK–STAT pathway, which can independent of a specific single mutation.

Patients with MF often, despite the presence of driver-type mutation, have additional mutations in genes responsible for epigenetic modulation. This includes genes responsible for posttranslational modification of histones (*ASXL1*, frequency 10–35%; *EZH2*, frequency 7–10%), DNA methylation (*TET2, DNMT3A, IDH1/2*), mRNA splicing (*SRFS2, SRF3B1*), and DNA repair (*TP53*). The presence of at least one mutation in *ASXL1, EZH2, SRSF2*, or *IDH1/2*, called high-molecular risk (HMR,) is associated with shorter OS and higher risk of blastic transformation [30]. Guglielmelli et al. [31] analysed the impact of mutations on ruxolitinib effectiveness in 166 patients from the COMFORT-II trial. No impact of mutations was seen on treatment effectiveness (defined as reduction of splenomegaly and/or systemic symptoms) and on haematological toxicity profile (including anaemia and thrombocytopaenia). The beneficial effect of ruxolitinib on OS was independent of mutations associated with poor prognosis. After a median observation of 151 weeks, the predicted survival of patients treated with ruxolitinib in week 144 was 0.79 in the HMR group and 0.85 in the low-molecular risk (LMR) group, compared with, respectively, 0.58 and 0.71 in patients receiving the best available therapy (BAT). Patel et al. [32] assessed the impact of mutations on spleen volume reduction and on time to treatment discontinuation (TTD) in 95 patients treated with ruxolitinib in phase I/II trials. The authors of the analysis found a significant, negative impact of the presence of at least one HMR mutation on splenic response. Additionally, patients in the HMR group were characterised by a shorter TTD and OS. Spiegel et al. [33] evaluated correlation between mutations and similar parameters in a cohort of 100 patients with MF treated with ruxolitinib (77 patients) or momelotinib (23 patients). Unlike the observation of Patel et al. [32], this analysis showed no correlation between the presence of mutations and splenic response. However, it confirmed the negative impact of mutations on time to treatment failure (TTF) and OS.

The results of the presented analysis indicate that the presence of mutation from the HMR group significantly impairs duration of response to ruxolitinib. Therefore, in patients with mutation, who are potential candidates for allo-HSCT, treatment with ruxolitinib should not postpone the decision regarding transplantation, and should be considered only as a part of preparation to the procedure.

Ruxolitinib and risk of venous and arterial embolisms

Chronic myeloproliferative neoplasms (MPN) are characterised by an increased risk of venous and arterial embolisms. This affects 10–30% of patients before and 10–20% after MPN diagnosis [34]. Thromboembolic disease is most common in patients with PV and less common in patients with essential thrombocytopaenia (ET) and MF [35]. Risk factors associated with an increased risk of venous and arterial embolisms are: prior history of thromboembolic disease, presence of *JAK2* V617F mutation, and leukocytosis over 15 G/l [36]. Pathogenesis of MPN-related embolisms is mostly based on disfunction of red blood cells, white blood cells, platelets, and epithelial cells that raises adhesion of blood cells to endothelium [36, 37]. *In vivo* studies of JAK2+ neutrocytes showed increased creation of neutrophil extracellular traps (NET), which play a crucial role in disposal of pathogens, immunological reactions, and clot development [38]. Patients with PV have additional risk factors due to the presence of rheologic disturbances associated with increased haematocrit.

Association between ruxolitinib and thromboembolic diseases in patients with MF and PV was found in a metanalysis that included data from 750 patients participating in the COMFORT-I, COMFORT-II (patients with MF), and RESPONSE (patients with PV) trials [39]. The authors concluded that treatment with ruxolitinib was associated with lower risk of arterial and venous embolic disease when compared to treatment with placebo or BAT.

Research from Italy assessed the effectiveness and safety of ruxolitinib in patients with MPN (12 patients with MF, five with PV, and four with ET), who had history of portal vein thrombosis. No aggravation of portal vein thrombosis or worsening of oesophageal varices was seen during ruxolitinib treatment. One haemorrhagic adverse event was reported [40].

Effectiveness and safety of ruxolitinib treatment in patients with PV refractory or intolerant to hydroxycarbamide was assessed in the RESPONSE (222 patients) [41]

and RESPONSE-2 (149 patients) [42] trials. Ruxolitinib was more effective than BAT, with a significantly lower rate of thromboembolic disease in patients receiving ruxolitinib.

One phase II trial compared the effectiveness and safety of ruxolitinib with BAT in patients with ET refractory or intolerant to hydroxycarbamide [43]. No difference between ruxolitinib and BAT was seen in response rate. After two years of treatment, no difference in rates of thromboembolic and haemorrhagic events was observed. Another trial evaluated the effectiveness of ruxolitinib as a second-line treatment in 39 patients with ET [44]. Thromboembolic disease was seen in two patients and non-significant bleeding events in four patients.

Concluding, available data suggest that treatment with ruxolitinib results a in lower rate of embolic disease in patients with MF and PV, without a similar effect seen in patients with ET. No increase in haemorrhagic events is seen in MPN patients receiving ruxolitinib.

How to withdraw ruxolitinib? Principles of ending therapy

According to the Polish National Health Fund Drug Program, ruxolitinib treatment should be stopped if there is no spleen size reduction is seen after three months of treatment and/or if the spleen size reduction is less than 50% as assessed in USG after six months of treatment. Other mentioned situations are the development of new or a clear increase in previously present systemic symptoms, as well as unacceptable toxicity despite proper dose reduction and/or introduction according to the Summary of Product Characteristics. The last indications for ruxolitinib withdrawal are loss of gained response (assessed every six months) and transformation into acute leukaemia.

The most common adverse events seen with ruxolitinib — dose-dependent anaemia and thrombocytopaenia — developed in, respectively, 40.4% and 44.5% of patients in the COMFORT-II trial and rarely caused ruxolitinib withdrawal [45]. Other common toxicities include: leukopaenia, diarrhoea, bleeding, infections, thromboembolic events, arterial hypertension, and elevated liver enzymes. The decision about stopping ruxolitinib should include the notion that adverse events are most common in the first six months of treatment and usually decrease thereafter [46].

Long-term observations indicate that ruxolitinib needs to be stopped in 55% of patients after three-year follow-up (data from COMFORT-I and -II trials) [46]. Median OS after stopping ruxolitinib is 14 months [47].

Severe adverse reactions after ruxolitinib withdrawal, called ruxolitinib distress syndrome (RDS), are described in the literature. As confirmed in clinical trials, benefit from ruxolitinib was associated with a significant decrease in serum pro-inflammatory cytokines, such as IL-6, IL-1RA, TNFα, macrophage inhibitory protein 1b (MIP-1b), or C-reactive protein (CRP) [48]. Therefore, RDS might be caused by a rapid increase in previously low cytokine concentration. Ruxolitinib distress syndrome includes various clinical manifestations, from brisk reoccurrence of disease-related symptoms (including fast increase of spleen size and development of cytopaenia) to more severe conditions such as acute respiratory distress syndrome (ARDS), disseminated intravascular coagulation (DIC), spleen infarction, tumour lysis-like syndrome, or tumour septic shock-like syndrome.

Luckily, RDS is very rare, and only 10 cases have been described in the literature so far. Tefferi et al. [49] described RDS in five out of 47 patients who finished ruxolitinib treatment. Among them, three developed ARDS, from who two required mechanical ventilation and catecholamine infusion due to septic shock-like syndrome; one other patient developed DIC-like syndrome. In the COMFORT-I trial one patient experienced fever, acute respiratory failure, and splenic haemorrhage with infarction [7]. Other literature reports describe tumour lysis-like syndrome [50], ARDS [51], and recurring respiratory failure that resolved each time ruxolitinib was re-initiated [52]. RDS can be diagnosed only after exclusion of other possible causes and no clinical, laboratory or pathology finding can be called pathognomonic. Time to RDS occurrence varies from less than 24 hours to more than three weeks after ruxolitinib withdrawal [48].

According to the available literature, management of RDS should include not only supportive care, antibiotics, and mechanical ventilation if necessary, but also steroids or re-introduction of ruxolitinib, which can be switched to other JAK2 inhibitors after achieving RDS remission. Because RDS is very rare, no data enable formulation of ruxolitinib withdrawal guidelines. The authors of RDS case reports suggest close observation, slow dose decrease, and concomitant introduction of steroids [48, 49].

Management of ruxolitinib-associated hyperleukocytosis

Leukocytosis can be found in CBC of about 10–25% of patients with MF [53]. An increase in leukocyte count to over 25 G/l is a poor prognostic factor included in the IPSS, DIPSS, and DIPSS Plus scales [20, 21, 23]. In most patients, ruxolitinib has little to no effect on leukocyte count. In the COMFORT-I trial, mean pre-treatment leukocytosis was between 20 and 30 G/l and decreased to 15–20 G/l during treatment [54]. Only few cases of

grade 3 and 4 leukopaenia, according to World Health Organisation classification, developed. Some patients may experience an increase in leukocytosis or even hyperleukocytosis (leukocyte count of > 50–100 G/l). Every such case should be evaluated for the rate of peripheral myeloblasts, to exclude MF transformation into leukaemia.

A drug that might be used to decrease the leukocyte count is hydroxyurea (HU) — a cytostatic drug commonly used in patients with MF [55]. Until today, there are only a limited number of reports describing concomitant usage of ruxolitinib and HU in patients with MF [56, 57]. Caocci et al. [56] reported a case of a female patient in whom ruxolitinib treatment resulted in a leukocyte count increase up to 94 G/l and subsequent reoccurrence of systemic symptoms. After initiation of HU at a daily dose of 500 mg, the leukocyte count returned to normal and systemic symptoms vanished. In another patient with hyperleukocytosis, combined ruxolitinib and HU treatment resulted in normalisation of leukocyte count, a decrease in spleen size, improvement in systemic symptoms, and lower blood transfusion dependency [57]. It seems that for patients who receive ruxolitinib and develop a significant rise in leukocyte count, HU might be a safe and efficient therapeutic option.

Metabolic disorders in patients receiving ruxolitinib

Metabolic disorders, such as decrease in body weight, low serum cholesterol and albumin concentration, or cachexia, are a common problem in patients with MF, especially in more advanced cases [58–60]. If present, they significantly impair the patient's prognosis [20, 23, 58–60]. The aetiology of metabolic disorders is multifactorial [4, 22, 23, 61–66]. On the one hand, massive splenomegaly can lead to abdominal symptoms (pain, nausea, vomiting, early satiety) and decrease appetite [4, 66]. On the other hand, aberration in JAK–STAT pathway signalling can lead to overproduction of pro-inflammatory cytokines such as IL-6 or TNFα, which induce chronic inflammation, hypercatabolic state, loss of body weight, induction of cachexia, and reduction of liver albumin production [62–64].

According to the results of two clinical trials, the COMFORT-I study (ruxolitinib *vs.* placebo) and the COMFORT-II study (ruxolitinib *vs.* BAT), ruxolitinib can efficiently inhibit JAK1 and JAK2 kinases, leading to spleen volume reduction (probably due to reduction of extra-medullar haematopoiesis), a decrease in systemic symptoms and improvement of quality-of-life in patients with MF. Reduction of pro-inflammatory cytokine concentration, TNFα, IL-6, and CRP might also play role [4, 65].

A gradual increase in body weight of patients receiving ruxolitinib has been noticed in the COMFORT-I, COMFORT-II, and COMFORT-III trials [7, 64, 66]. This observation was confirmed in *post hoc* analysis of long-term (96 weeks) data from the COMFORT-I trial [61]. Among patients receiving ruxolitinib, 96.1% of patients achieved any body weight increase (mean 3.9 kg, as opposed to mean loss of 1.9 kg in patients receiving placebo; $p < 0{,}0001$) after 24 weeks of therapy, with comparable results after 36 weeks and even more profound gain of a mean 5.7 kg after 96 weeks. Body mass index (BMI) analysis showed a significant mean gain of 1.4 kg/m^2 after 24 weeks in the ruxolitinib arm (compared with a mean 0.7 kg/m^2 loss in the placebo arm; $p < 0.0001$), with comparable results after 36 weeks of treatment.

Additionally, the COMFORT-I trial also evaluated the concentration of leptin as a marker of adipose tissue. In patients receiving ruxolitinib, a more than two-fold increase in mean plasma leptin concentration was noted after four weeks of treatment and remained significant after 24 weeks. In patients receiving placebo a slight decrease in leptin concentration was noted during the same observation period [65].

In a *post hoc* analysis of the COMFORT-I trial, a rise in cholesterol concentration was noted in 96.8% of patients receiving ruxolitinib. After 24 weeks cholesterol increased 26.4% from baseline (29.5 mg/dl) in patients receiving ruxolitinib compared to a 3.3% fall (4.98 mg/dl) in patients receiving placebo. The cholesterol increase in the ruxolitinib group was maintained after 96 weeks of therapy (35.8% increase from baseline, 38 mg/dl). It should be emphasised that cholesterol concentration did not exceed 240 mg/dl of complete cholesterol and 160 mg/dl of low-density lipoprotein (LDL) cholesterol, thus not resulting in a higher risk of hypercholesterolaemia [61].

As with cholesterol, *post hoc* analysis of COMFORT-I data showed that 94.8% of patients receiving ruxolitinib experienced a rise in albumin concentration. The increase reached 5.8% (2.3 g/dl) at week 24 (compared to 1.7% [0.8 g/dl] decrease with placebo; $p < 0.0001$), with a stable results at week 10 and an additional rise of 7.6% (3.1 g/dl) at week 96 [61].

Both body mass increase and rise of cholesterol and albumin concentration was independent of the degree of spleen size reduction (≥ 35% *vs.* 10–35% *vs.* < 10%) and of the degree of systemic symptom reduction assessed with MyeloProliferative Neoplasm — Total Symptom Score (MPN-TSS) (not less than 50% *vs.* less than 50%) [61].

To sum up, ruxolitinib treatment, through inhibition of JAK1 and JAK2 kinases, leads to a decrease of pro-inflammatory cytokines concentration (IL-6 and TNFα). This results in reduction of chronic inflamma-

tion and systemic symptoms, suppression of hypercatabolism, and decrease in spleen size. Secondary to this, but no less important, is the observation that patients receiving ruxolitinib experience gradual and consistent (for over 96 weeks) improvement in body weight, cholesterol concentration (without increased risk of hypercholesterolaemia and cardio-vascular disorders), and albumin concentration. This effect might be partially responsible for the survival benefit associated with ruxolitinib in this patient population. Close follow-up of nutritional markers might provide valuable insights during ruxolitinib therapy.

Conclusions

The presented contemplations regarding clinical aspects of ruxolitinib treatment in patients with MF should provide answers to basic questions and doubts that may arise during therapy. The discussed issues concentrate mostly on management of patients with distinctive clinical and/or pathological profile and on dealing with certain adverse events. Crucial value can be attributed to proper monitoring of systemic symptoms, which are the best indicators of MF activity and can also overlap with possible adverse events.

The most important symptoms of MF are splenomegaly and cytokine-induced systemic symptoms that include weight loss, night sweats, fatigue, fever, and pruritus. Because the presence of systemic symptoms is required to qualify patients to the Polish National Health Fund Drug Program, and subsequent changes in symptoms provide insight into treatment effectiveness, objective symptom evaluation is crucial. This can be achieved with the MPN-TSS scale, which includes: fatigue; early satiety; discomfort in abdomen; decrease in activity and concentration; night sweats; pruritus; bone pains; fever; and unintentional weight loss.

Other important issue includes ruxolitinib distress syndrome, which can arise when ruxolitinib is withdrawn rapidly. Because this may lead to fierce and symptomatic cytokine storm, ruxolitinib withdrawal should be a gradual process. Nonetheless, stopping ruxolitinib may result in reoccurrence of systemic symptoms and an increase in spleen size. The decision regarding ruxolitinib withdrawal should be taken after careful deliberation and should be properly planned to limit the possibility of unexpected complications.

Patients with MF receiving ruxolitinib should be closely monitored to detect both haematological and non-haematological adverse events. The most common haematological adverse events are anaemia and thrombocytopaenia. In the COMFORT-I and COMFORT-II trials, all-grade anaemia according to CTCAE occurred in nearly all treated patients, with grade 3 and 4 events in 45.2% of patients in the COMFORT-I and 62% of patients in the COMFORT-II trial. Anaemia usually develops within the first eight weeks of treatment, with the nadir of Hb concentration between weeks 8 and 12, and then gradually increases and stabilises after six months of therapy. More than 50% of patients require blood transfusions, but ruxolitinib-related anaemia rarely requires dose modification or interruption. Other causes of anaemia should be ruled out, just as progression of MF itself.

All grade thrombocytopaenia, according to CTCAE, occurred in 70% of patients in the COMFORT-I and COMFORT-II trials. About 11% of patients experienced grade 3 and 4 thrombocytopaenia. Median time to thrombocytopaenia development was about eight weeks. Thrombocytopaenia was reversible after dose reduction or drug interruption, with a median time to PLT count recovery to over 50 G/l of two weeks. Decrease in PLT count might require ruxolitinib dose adjustment, mostly to avoid any treatment interruption that may limit therapy effectiveness. Patients with PLT count lower than 50 G/l should not be qualified for treatment with ruxolitinib. An additional indication for treatment interruption is neutropaenia greater than 0.5 G/l.

Regardless of PLT count, ruxolitinib treatment often results in haemorrhagic adverse events, most commonly subcutaneous haemorrhages, occurring in about 20% of patients. In the COMFORT-I and COMFORT-II trials, gastrointestinal bleeding of all grades occurred in 5% of patients and grade 3 and 4 events in 1.3% of patients. Intracranial bleeding developed in 1% of patients. Other bleeding events (including nosebleed, haematuria, or procedural bleedings) of all grade occurred in 13% of patients and of grade 3 and 4 in 2.3% of patients.

Non-haematological adverse events associated with ruxolitinib include headaches, dizziness (in about 15% of patients), diarrhoea, and mild to moderate increase in AlAt and AspAT activity (in about 20% of patients). Additionally, as a result of reduction of pro-inflammatory cytokine concentration, ruxolitinib exhibits immunosuppressive properties, including inhibition of dendritic cell activity, which leads to the suppression of CD4+ and CD8+ lymphocytes. Consequently, patients treated with ruxolitinib are more prone to infections, including opportunistic ones. Data from the COMFORT trials show increased risk of urinary tract infections and *Herpes zoster* infections in patients treated with ruxolitinib. Cases of HBV reactivation, tuberculosis, *Cryptococcus neoformans* pneumonia, toxoplasmosis uveitis, and progressive multifocal leukoencephalopathy were also reported. Therefore, screening for tuberculosis and hepatotropic viruses as part of routine pre-treatment evaluation should be considered. If positive, proper prophylaxis should be undertaken.

Ruxolitinib is excreted through kidneys and, to a lesser degree, through the digestive tract. Patients with severe renal impairment should receive reduced initial dose and patients with end-stage renal failure undergoing dialysis should receive a single daily dose after each dialysis. Patients with impaired liver function should receive 50% of the standard dose.

Because ruxolitinib interacts with numerous other agents, simultaneously used drugs should be revised and the ruxolitinib dose reduced if necessary. This is mostly due to the ruxolitinib metabolism, which involves mainly cytochrome CYP3A4 and partially CYP2C9. Fluconazole, a strong inhibitor of both mentioned cytochromes, increases ruxolitinib serum concentration by 100–300%. Therefore, the ruxolitinib dose should be reduced by 50%, with the same dosing schedule, if strong CYP3A4 inhibitors are administered (antifungal agents such as fluconazole, ketoconazole, itraconazole, posaconazole, voriconazole; antiviral agents such as boceprevir, ritonavir, nelfinavir; and antibacterial agents such as clarithromycin).

Patients receiving ruxolitinib simultaneously with CYP3A4 inducers (such as carbamazepine, phenobarbital, phenytoin, rifampicin, St. John's wort) should be carefully monitored and ruxolitinib dose increased according to achieved effectiveness and safety. No ruxolitinib dose adjustment is required when combined with mild and moderate CYP3A4 inducers (such as ciprofloxacin, erythromycin, diltiazem, cimetidine, atazanavir, and amprenavir).

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