A case report of cryptococcal meningitis associated with ruxolitinib

Daisuke Tsukui, MD, Hiroaki Fujita, MD, PhD*, Keisuke Suzuki, MD, PhD, Koichi Hirata, MD, PhD

Abstract
We herein report a 76-year-old Japanese man with myelofibrosis who developed cryptococcal meningitis. After treatment for 5 months with ruxolitinib, the patient presented with fever and disturbance of consciousness. Marked nuchal stiffness was noted. The magnetic resonance imaging results of the brain were normal. Lumbar puncture showed an opening cerebrospinal fluid (CSF) pressure of 110 mm H2O, pleocytosis (85 mononuclear cells and 222 polymorphonuclear cells/μL; elevated protein (194 mg/dL), Blood and CSF cultures grew no bacteria or fungi. However, cryptococcal antigen was detected in the blood and CSF samples. We discontinued ruxolitinib and started administration of amphotericin B. His condition improved gradually 1 week after initiation of treatment. There have been only a few reports on cryptococcal meningitis associated with ruxolitinib. Physicians should consider the possibility of cryptococcal meningitis in patients receiving ruxolitinib.

Abbreviations: CSF = cerebrospinal fluid, DCs = dendritic cells, JAK = Janus kinase, MF = myelofibrosis, MPL = myeloproliferative leukemia virus oncogene, NK = natural killer, PV = polycythemia vera, STAT = signal transducer and activator of transcription.

Keywords: cryptococcus, immunosuppression, Janus kinase inhibitor, meningitis, ruxolitinib

1. Introduction
Ruxolitinib, an inhibitor of Janus kinase (JAK) 1 and 2, has been approved for the treatment of myelofibrosis (MF) and polycythemia vera (PV) by reducing spleen size, ameliorating debilitating symptoms, and improving overall survival.[1,2] The JAK/signal transducer and activator of transcription (STAT) pathway is the principal signaling mechanism for numerous cytokines and growth factors. JAK inhibitors exert immunosuppressive activities through the downregulation of several cytokines, such as interleukins, interferon-γ, and tumor necrosis factor-α,[3] and result in dysfunction of dendritic cells (DCs),[4] T-regulatory cells,[5] and natural killer (NK) cells.[6] Cryptococcal meningitis is known to occur particularly frequently in immunocompromised hosts.[7] However, there have been only 2 reports of cryptococcal meningitis in patients treated with JAK inhibitors. Here, we report a case of cryptococcal meningitis in a ruxolitinib-treated patient with primary MF.

2. Case report
At the age of 64 years, the present patient was diagnosed with essential thrombocythemia, and hydroxycarbamide was initiated. His condition had been stable for several years. At the age of 72 years, anagrelide hydrochloride hydrate was started instead of hydroxycarbamide because of worsening thrombocytosis. In February 2019, the patient was diagnosed with MF according to the results of bone-marrow puncture. Although the JAK2 mutation was negative, the myeloproliferative leukemia virus oncogene (MPL) W515L mutation was detected. Since then, he had been treated with ruxolitinib (10 mg/d). At the age of 76 years (5 months after initiation of ruxolitinib), the patient was admitted to our hospital because of high-grade fever and disturbance of consciousness from a day before admission. On examination, his body temperature was 38.8°C; his other vital signs were normal. Marked nuchal stiffness was noted. The patient was disoriented to time and place. Cranial nerves were intact. There was no motor weakness or cerebellar ataxia. Tendon reflexes were normal and symmetrical without any pathological reflexes. No sensory impairment was noted. Laboratory data showed mildly elevated C-reactive protein levels (0.31 mg/dL) and procalcitonin levels (0.10 ng/mL). Markedly elevated ferritin levels (2203.5 ng/mL) were observed. The white blood cell count (7000/µL) and platelet count (25.9 × 10^9/µL) were preserved, but the red blood cell count was decreased (227/µL). Normal levels of β-d-glucan were observed (6.0 pg/mL). Lumbar puncture yielded an opening cerebrospinal fluid (CSF) pressure of 110 mm H2O and pleocytosis (85 mononuclear cells and 222 polymorphonuclear cells/μL). The CSF glucose level was 69 mg/dL with a low CSF/serum glucose ratio of 43%, and the protein level (194 mg/dL) was elevated. Herpes simplex virus DNA and varicella-zoster virus DNA were negative. CSF cultures grew no bacteria or fungi. A human immunodeficiency virus test was negative. The

Received: 17 December 2019 / Received in final form: 4 February 2020 / Accepted: 18 February 2020
http://dx.doi.org/10.1097/MD.0000000000019587
magnetic resonance imaging results of the brain were normal. Figure 1 shows the clinical course and treatment of the patient. He was suspected of having meningitis and was empirically treated with micafungin (150mg/d) and cefozopran (4g/d), followed by meropenem (3g/d), acyclovir (2250mg/d), and amphotericin B (350mg/d). However, cryptococcal antigen was detected in CSF (titers, 1:16) and serum on day 6 (Table 1). We discontinued the treatment with ruxolitinib and continued the administration of amphotericin B (350mg/d), and the patient’s condition improved until day 10. Amphotericin B was used until day 37, followed by administration of fluconazole (400mg/d). The patient was on continuous therapy at the time of this report.

3. Discussion

Ruxolitinib is a selective JAK 1/2 inhibitor that has been approved for the treatment of MF and PV. The JAK/STAT pathway plays an important role in hematopoiesis and the immune response in vivo. After engagement of the receptor by the corresponding ligand, JAK becomes activated via phosphorylation, followed by JAK/STAT pathway activation. Activated STATs dimerize and translocate to the nucleus, where they regulate transcription and release proinflammatory cytokines and growth factors, including erythropoietin, granulocyte macrophage colony-stimulating factor, and thrombopoietin. In patients with MF, gene mutations, such as those in JAK2 and MPL, are in a constant phosphorylated state, independent of the binding of ligand to its receptor. Excess release of proinflammatory cytokines and growth factors triggers the systemic symptoms of MF and ineffective hematopoiesis. Blockage of JAK1 mainly improves systemic symptoms via a reduction in proinflammatory cytokines, and blockage of JAK2 mainly improves splenomegaly and anemia via a reduction in growth factors and prevents ineffective hematopoiesis. However, some opportunistic infections related to ruxolitinib have been reported previously.

The present case involved cryptococcal meningitis in a patient treated with ruxolitinib. To the best of our knowledge, only 2 cases of cryptococcal meningitis in ruxolitinib-treated patients have been previously reported. Table 2 summarizes cryptococcal meningitis associated with ruxolitinib, including our case, and 2 clinical features were found. First, the opening pressure of lumbar puncture in the present case was not high, although raised CSF pressure is one of the typical clinical features of cryptococcal meningitis. Half of patients with cryptococcal meningitis show a CSF opening pressure over 250mm H₂O; additionally, a quarter of patients show an extremely high pressure over 350mm H₂O. The mechanism of high CSF pressure is presumed to block CSF reabsorption by live or dead organisms, with shed cryptococcal polysaccharide at the level of the arachnoid granulations and other CSF reabsorption sites. Loyse et al. reported that arachnoid granulation tissue contains many fungal cells in comparison with other sites of the brain, and high numbers of organisms are associated with increased antemortem CSF pressure. Bicanic et al. reported that high CSF pressure in cryptococcal meningitis is associated with the phenotype of an infectious Cryptococcus neoformans strain and host factors other than the numbers of fungal cells. Among 3 patients with cryptococcal meningitis associated with ruxolitinib, 2 had normal opening CSF pressure (opening CSF pressure was not describe in 1 patient) (Table 2). Among the 3 cases, no trend was observed in CSF findings, such as the degree of pleocytosis and protein elevation. To the best of our knowledge, there has been no report of cryptococcal meningitis associated with ruxolitinib showing the numbers of fungal bodies in a postmortem study. Because only 2 cases have previously been reported, more studies are needed to confirm whether a normal CSF pressure is one of the features of cryptococcal meningitis associated with ruxolitinib or just the finding in our case.

Second, the outcome of the present case was relatively good compared to typical cryptococcal meningitis. The 2 previous cases of cryptococcal meningitis associated with ruxolitinib also
IgG 839 mg/dL (861, C4, 48.4 mg/dL (11.0
Ferritin 2203.5 ng/mL (21.8
Cryptococcus
CMV antigen; negative Quantitative assay of
antigen 0.1 (0.5
Tb interferon-
C-reactive protein 0.31 mg/dL (0.14
Creatine kinase 150 IU/L (59
Creatinine 0.91 mg/dL (0.65
Uric acid 4.1 mg/dL (3.7
K 4.7 mmol/L (3.6
Cl 101 mmol/L (101
ACE 8.2 IU/L (8.3
Prakash et al, 2019 51 males PV Cryptococcal meningitis

Table 2
Cryptococcal meningitis associated with ruxolitinib.

| Age/sex Underlying disease | Interval, amount | CSF pressure | CSF findings | WBC count at onset | Treatment | Outcome |
|-----------------------------|------------------|--------------|--------------|------------------|-----------|---------|
| Prakash et al, 2019 51 males PV Cryptococcal meningitis disseminated histoplasmosis | 18 months NR | Cell 19/µL TP 72 mg/dL Glu 27 mg/dL | 8002/µL L-AMB, 5-FC→ isavuconazole | Survived |
| Chen et al, 2016 69 females MF Cryptococcal meningoencephalitis | 46 months 20 mg/d | 140 mm H2O | Cell 420/µL TP 108 mg/d Glu normal | NR FLCZ, L-AMB | Survived |
| Present case 76 males MF Cryptococcal meningitis | 5 months 10 mg/d | 110 mm H2O Cell 307/µL TP 194 mg/dL Glu 69 mg/dL | 8100/µL MCFG→L-AMB | Survived |

5-FC = flucytosine, FLCZ = flocanazole, Glu = glucose, Interval = between began to use ruxolitinib and onset of meningitis, L-AMB = amphotericin, MF = myelofibrosis, NR = not reported, PV = polycythemia vera, TP = total protein.

showed good clinical outcomes (Table 2). In the present case, ruxolitinib was discontinued on day 6, and in 1 previous report, ruxolitinib was discontinued when fungal infection was found (in the other case, whether ruxolitinib was discontinued was not described). Hirano et al suggested that ruxolitinib administration should be discontinued if possible; otherwise, the treatment with ruxolitinib may be ineffective against a pulmonary cryptococcosis infection. Additionally, in our case, discontinuing ruxolitinib may lead to a good outcome; therefore, as ruxolitinib may impact the immune response against cryptococcosis, ruxolitinib should be discontinued immediately when cryptococcal meningitis is suspected.

A phase III study of ruxolitinib reported that reactivation of tuberculosis and herpes zoster virus were the predominant opportunistic infections observed with ruxolitinib. Since that time, some cases of opportunistic infection associated with ruxolitinib have been reported. Dioveriti et al published a review of 32 cases identified as opportunistic infections associated with ruxolitinib. Although the majority of cases reported were reactivations of tuberculosis (34%), several fungal infections were also reported (22%), and cryptococcosis was the most frequently reported fungus. In a phase II, phase III, and long-term extension clinical trial with 5671 patients treated with tofacitinib, another JAK inhibitor approved for the treatment of adult patients with rheumatoid arthritis, cryptococcal infections were also reported (2 pulmonary infections and 1 case of meningitis).

Many studies have tried to elucidate the mechanism by which ruxolitinib impacts the immune system. A review of the literature conducted by Manduzio indicated that the immunological derangement of ruxolitinib is mainly based on T cells, DCs, and NK cell defects. Heine et al reported that ruxolitinib affects the function and phenotype of DCs, leading to impaired T-cell activation. Ostoji et al reported that ruxolitinib suppresses cell-mediated immunity by inhibiting the T-helper lymphocyte 1 response and reducing the production of interferon-γ. The host defense against C. neoformans is associated with cell-mediated immunity, especially accomplished by the combined action of activated macrophages, NK cells, and T cells. In addition, Hardison et al reported that STAT1 and signaling through the JAK/STAT pathway play an important role in the protective response against cryptococcosis via STAT1-mediated classical macrophage activation. In the present case, suppression of anticytotoxic responses was likely to induce the development of cryptococcal meningitis, as in the previously reported cases of cryptococcal infection. Ruxolitinib-associated opportunistic infections are not time-dependent and may occur any time after initiation of the drug. However, whether this effect is dose dependent is still controversial.
In conclusion, we report a case of cryptococcal meningitis associated with ruxolitinib. Ruxolitinib administration is known to lead to opportunistic infections, and thus, it can cause cryptococcal meningitis, as the incidence of cryptococcal meningitis increases in patients with immunosuppressant conditions. Physicians should consider the possibility of cryptococcal meningitis in patients receiving ruxolitinib and discontinue the drug if possible when high-grade fever persists even in the absence of headache. Because ruxolitinib is a relatively new drug, further accumulation of clinical experience to monitor possible side effects is needed.

Acknowledgment

The authors thank Dr Honoka Arai and Dr Yuiko Nakamura, Department of Hematology and Oncology, Dokkyo Medical University Hospital, for their assistance with this study.

Author contributions

Conceptualization: Daisuke Tsukui, Hiroaki Fujita. Writing – original draft: Hiroaki Fujita, Keisuke Suzuki. Writing – review & editing: Hiroaki Fujita, Koichi Hirata. Hiroaki Fujita orcid: 0000-0003-2184-7916.

References

[1] Harrison C, Kiladjian JJ, Al-Ali HK, et al. JAK inhibition with ruxolitinib versus best available therapy for myelofibrosis. N Engl J Med 2012;366:787–98.
[2] Verstovsek S, Mesa RA, Gotlib J, et al. A double-blind, placebo-controlled trial of ruxolitinib for myelofibrosis. N Engl J Med 2012;366:799–807.
[3] Quintas-Cardama A, Verstovsek S. Molecular pathways: Jak/STAT pathway: mutations, inhibitors, and resistance. Clin Cancer Res 2013;19:1933–40.
[4] Heme A, Held SA, Daecke SN, et al. The JAK-inhibitor ruxolitinib impairs dendritic cell function in vitro and in vivo. Blood 2013;122:1192–202.
[5] Parampalli Yajnanarayana S, Stubig T, Cornez I, et al. JAK1/2 inhibition impairs T cell function in vitro and in patients with myeloproliferative neoplasms. Br J Haematol 2015;169:924–33.
[6] Schonberg K, Rudolph J, Vonnahme M, et al. JAK inhibition impairs NK cell function in myeloproliferative neoplasms. Cancer Res 2015;75:2187–99.
[7] Williamson PR, Jarvis JN, Panackal AA, et al. Cryptococcal meningitis: epidemiology, immunology, diagnosis and therapy. Nat Rev Neurol 2017;13:15–24.
[8] Rawlings JS, Rosler KM, Harrison DA. The JAK/STAT signaling pathway. J Cell Sci 2004;117(Pt 8):1281–3.
[9] Murray PJ. The JAK-STAT signaling pathway: input and output integration. J Immunol 2007;178:2623–9.
[10] Pikman Y, Lee BH, Mercher T, et al. MPLW515L is a novel somatic activating mutation in myelofibrosis with myeloid metaplasia. PLoS Med 2006;3:e270.
[11] Verstovsek S. Therapeutic potential of Janus-activated kinase-2 inhibitors for the management of myeloproliferative neoplasms. Clin Cancer Res 2010;16:1988–96.
[12] Vannucchi AM, Guglielmelli P, Tefferi A. Advances in understanding and management of myeloproliferative neoplasms. CA Cancer J Clin 2009;59:171–91.
[13] Quintas-Cardama A, Vaddi K, Liu P, et al. Preclinical characterization of the selective JAK1/2 inhibitor INCB018424: therapeutic implications for the treatment of myeloproliferative neoplasms. Blood 2010;115:3109–17.
[14] Cazzola M. Somatic mutations of JAK2 exon 12 as a molecular basis of erythrocytosis. Haematologica 2007;92:1583–9.
[15] Tefferi A, Vaidya R, Caramazza D, et al. Circulating interleukin (IL)-8, IL-2R, IL-12, and IL-15 levels are independently prognostic in primary myelofibrosis: a comprehensive cytokine profiling study. J Clin Oncol 2011;29:1356–63.
[16] Prakash K, Richman D. A case report of disseminated histoplasmosis and concurrent cryptococcal meningitis in a patient treated with ruxolitinib. BMC Infect Dis 2019;19:287.
[17] Chen CC, Chen YY, Huang CE. Cryptococcal meningoen cephalitis associated with the long-term use of ruxolitinib. Ann Hematol 2016;95:361–2.
[18] Hirano A, Yamasaki M, Saito N, et al. Pulmonary cryptococcosis in a ruxolitinib-treated patient with primary myelofibrosis. Respir Med Case Rep 2017;22:87–90.
[19] Liu J, Mouhayer E, Tarrand JJ, et al. Fulminant Cryptococcus neoformans infection with fatal pericardial tamponade in a patient with chronic myelomonocytic leukaemia who was treated with ruxolitinib: case report and review of fungal pericardi tis. Mycoses 2018;61:245–55.
[20] Goldberg RA, Reichel E, Oshey LJ. Bilateral toxoplasmal reinitis associated with ruxolitinib. N Engl J Med 2013;369:681–3.
[21] Lee SC, Feenstra J, Georgiou PR. Pneumocystis jirovecii pneumonitis complicating ruxolitinib therapy. BMJ Case Rep 2014;2014:
[22] Wysham NG, Sullivan DR, Allada G. An opportunistic infection associated with ruxolitinib, a novel janus kinase 1,2 inhibitor. Chest 2013;143:1478–9.
[23] Wathes R, Moule S, Milojkovic D. Progressive multifocal leukencephalopathy associated with ruxolitinib. N Engl J Med 2013;369:197–8.
[24] Colomba C, Rubino R, Siracusa L, et al. Disseminated tuberculosis in a patient treated with a JAK2 selective inhibitor: a case report. BMC Res Notes 2012;5:552.
[25] Shen CH, Hwang CE, Chen YY, et al. Hepatitis B virus reactivation associated with ruxolitinib. Ann Hematol 2014;93:1075–6.
[26] Loyse A, Wainwright H, Jarvis JN, et al. Histopathology of the arachnoid granulations and brain in HIV-associated cryptococcal meningitis: correlation with cerebrospinal fluid pressure. AIDS 2010;24:405–10.
[27] Bicanic T, Brouwer AE, Meintjes G, et al. Relationship of cerebrospinal fluid pressure, fungal burden and outcome in patients with cryptococcal meningitis undergoing serial lumbar punctures. AIDS 2009;23:701–6.
[28] Harrison CN, Vannucchi AM, Kiladjian JJ, et al. Long-term findings from COMFORT-II, a phase 3 study of ruxolitinib vs best available therapy for myelofibrosis. Leukemia 2016;30:1701–7.
[29] Dioverti MV, Abu Saleh OM, Tande AJ. Infectious complications in patients on treatment with ruxolitinib: case report and review of the literature. Infect Dis (Lond) 2018;50:381–7.
[30] Harigai M. Growing evidence of the safety of JAK inhibitors in patients with chronic myelomonocytic leukaemia who was treated with ruxolitinib: case report and review of fungal pericarditis. Mycoses 2018;61:245–55.
[31] O'Brien S, Seligson D, Butturini A, et al. Ruxolitinib for the treatment of myelofibrosis: to be or not to be an immunosuppressant. Haematologica 2007;92:1595–7.
[32] Kronstad J, Saikia S, Nielson ED, et al. Adaptation of the selective JAK1/2 inhibitor INCB018424: therapeutic implications for the treatment of myeloproliferative neoplasms. Blood 2010;115:3109–17.
[33] Ostojic A, Vrhovac R, Verstovsek S. Ruxolitinib for the treatment of chronic myelomonocytic leukaemia: a phase 3 study of ruxolitinib vs best available therapy for myelofibrosis. Leukemia 2015;29:1356–63.
[34] Hardison SE, Herrera G, Young ML, et al. Protective immunity against hepatitis B virus infection is impaired by ruxolitinib. J Clin Oncol 2018;36:2123–30.
[35] Heine A, Held SA, Daecke SN, et al. The JAK-inhibitor ruxolitinib impairs dendritic cell function in vitro and in vivo. Blood 2013;122:1192–202.