Long-Term Response to Rituximab in Patients with Relapsing Thrombotic Thrombocytopenic Purpura

Gideon Y. Stein MD[1], Dorit Blickstein MD[2], Jerome Orlin MD[3], Galit Sarig MD[4] and Aida Inbal MD[2]

[1]Department of Internal Medicine B, [2]Thrombosis and Hemostasis Unit and Hematology Institute, and [3]Department of Transfusion Medicine, Rabin Medical Center (Beilinson Campus), Petah Tikva, affiliated with Sackler Faculty of Medicine, Tel Aviv University, Ramat Aviv, Israel
[4]Hematology Laboratory, Thrombosis and Hemostasis Unit, Rambam Health Care Campus, Haifa, Israel

ABSTRACT: Acquired thrombotic thrombocytopenic purpura (TTP) is an uncommon disease in adults, characterized by fever, neurological manifestations, microangiopathic hemolytic anemia, thrombocytopenia, renal dysfunction, and the presence of antibodies against the enzyme ADAMTS13. Treatment with plasmapheresis has increased the survival from 10% to more than 90%. Still, there is a subset of patients with resistant TTP who fail to respond to plasmapheresis or remain dependent on this procedure. There is mounting evidence that rituximab may play an important role in remission induction of resistant/relapsing TTP, but the extent of the remission is unknown. We present here four patients with chronic-relapsing TTP who responded favorably to rituximab. All four patients achieved prolonged remission of 23 to 82 months after the treatment. One patient relapsed 6 years after the initial treatment with rituximab and re-entered remission following retreatment.

IMAJ 2011; 13: 398–401

KEY WORDS: thrombotic thrombocytopenic purpura, ADAMTS13, rituximab

For Editorial see page 436

Thrombotic thrombocytopenic purpura is characterized mainly by the presence of an arteriolar microangiopathy including platelet thrombi that ultimately leads to thrombocytopenia and microangiopathic hemolytic anemia. Multiple studies have shown that very large circulating von Willebrand factor multimers are responsible for TTP in adults [1]. vWF multimers are usually cleaved by ADAMTS13 (vWF, cleaving protease) [1,2]. In patients with TTP, autoantibodies have been found that inhibit ADAMTS13, resulting in inhibition of the cleavage of these large vWF multimers [2] which have a particularly high affinity to platelet glycoprotein receptors [3]. The presence of these high molecular weight multimers is consistent with increased platelet adhesion and thrombus formation in small blood vessels [4].

Plasmapheresis using fresh frozen plasma has become the standard treatment for TTP. Most patients respond to this treatment and achieve remission. Some patients, however, experience multiple relapses, developing a chronic debilitating form of the disease that can be life threatening, or they become plasmapheresis-dependent [5,6]. Recently, a number of reports have suggested an important role for rituximab in the treatment of resistant/relapsing acquired TTP, especially in patients with anti-ADAMTS13 antibodies [7,8]. It is assumed that by eliminating B cells, rituximab also eliminates autoantibody-secreting B cells, which are associated with TTP and other autoantibody-related diseases. In the absence of anti-ADAMTS-13 antibodies, high molecular weight vWF multimers are cleaved, while the remaining multimers no longer aggregate platelets to form arterial thrombi.

Although the role of rituximab in remission induction of relapsing TTP has been recognized, the length of the remission is unknown. We report on four patients with chronic-relapsing TTP treated with rituximab. All patients achieved prolonged remissions following treatment.

TTP = thrombotic thrombocytopenic purpura
vWF = von Willebrand factor

PATIENTS AND METHODS

The diagnosis of TTP was established by the detection of microangiopathic anemia on blood film, thrombocytopenia and the presence of anti-ADAMTS13 antibodies (in some patients). All other reasons for thrombocytopenia or hemolytic anemia were ruled out. Laboratory and clinical data are summarized in Table 1.

PATIENT 1

A 55 year old woman presented in 2007 with a non-traumatic epidural hematoma and seizures. Treatment with corticosteroids and plasmapheresis resulted in a complete remission. During the ensuing 3 months two asymptomatic relapses were diagnosed during routine follow-up. Both were successfully treated with plasmapheresis and corticosteroids. Due to high operative risk, splenectomy was not performed. Because of her history of

Table 1. Patient characteristics

Patient #	Gender	Age at presentation (yrs)	Plt (10^9/L)	Hb (g/dl)	LDH (units/L)	MAHA	ADAMTS13 activity (%)*	Anti-ADAMTS13 Ab (U/ml)**	TTP episodes	Treatment modalities	Rituximab	Response
1	F	55	20	7.0	1873	+	N/A	N/A	3	Steroids, plasmapheresis	600 mg/ wk/4 wks	Complete remission (3 yrs, 8 mos)
2	M	37	13	15.4	2574	+	N/A	N/A	3	Steroids, plasmapheresis	700 mg/ wk/4 wks	Complete remission (1 yr, 11 mos)
3	M	41	7	8.7	3700	+	15	42	6	Steroids, plasmapheresis, splenectomy	670 mg/ wk/4 wks	Complete remission (3 yrs, 6 mos)
4	F	26	13	7.1	4259	+	‹ 1.0	100	4	Steroids, plasmapheresis, splenectomy, vincristine	500 mg/ wk/4 wks	Relapse after 6 years, complete remission after re-treatment, 19 mos remission

*Normal range 65–120%
**Normal range 0–15 U/ml
Plt = platelets, LDH = lactate dehydrogenase, TTP = thrombotic thrombocytopenic purpura, MAHA = microangiopathic hemolytic anemia, N/A = not available

only short-lived remissions, she was treated with rituximab and achieved complete remission lasting more than 44 months.

PATIENT 2

A 37 year old man presented in 2008 with abdominal pain and nephritic syndrome. Plasmapheresis resulted in a complete remission. A relapse 9 months later was treated with plasmapheresis, however without response. Administration of rituximab resulted in complete remission, lasting over 23 months. It is worth mentioning that following rituximab treatment the nephritic syndrome also resolved.

PATIENT 3

A 41 year old man presented in 1993 with fever, hemiparesis, purpura and hematuria. Treatment with plasmapheresis resulted in complete remission. After five relapses, which were successfully treated with plasmapheresis, a sixth relapse, 6 years after the initial presentation, failed to respond to plasmapheresis. Remission was achieved only after splenectomy and treatment with high-dose steroids, intravenous immunoblobulin and vincristine. In the following years the disease entered a chronic-relapsing phase and was treated with therapeutic/prophylactic plasmapheresis every few weeks. In 2007, laboratory testing revealed a high titer of anti-ADAMTS13 antibodies and low activity of ADAMTS13. Rituximab was administered in 2008, and a complete remission, lasting more than 42 months, was achieved. Interestingly, although the patient was asymptomatic, ADAMTS13 activity has remained persistently low even after treatment with rituximab.

PATIENT 4

A 23 year old woman presented in 1992 with weakness, low back pain and confusion. Plasmapheresis resulted in a complete remission. A year later, a relapse was treated by splenectomy, resulting in a disease-free interval of 5 years. In 1998 she experienced a severe relapse, manifested by hemolytic anemia, thrombocytopenia, renal failure and neurological complications including confusion, aphasia and coma with respiratory arrest, She was treated with plasmapheresis, corticosteroids, IVIG, vincristine and supportive care. A complete clinical and laboratory remission was achieved again, as published in a previous report [9]. In 2003 the patient experienced another severe relapse, manifested by epistaxis, hemoptysis and macrohematuria. She again responded to treatment with plasmapheresis and corticosteroids; however, all attempts to taper plasmapheresis resulted in relapse. After 2 months of intensive treatment that was complicated by sepsis and seizures, the patient received a single dose of cyclophosphamide, 1.2 g, and four weekly courses of rituximab [Table 1]. For the next 6 years, the patient remained in remission, as reported previously by some of us [10]. In November 2009, another relapse occurred that manifested as generalized weakness, epistaxis, hematuria and severe thrombocytopenia. High levels of anti-ADAMTS13 antibodies were detected in the patient's plasma, accompanied by undetectable levels of ADAMTS13 activity. The patient had been previously exposed to hepatitis B virus, with positive anti-HBc and anti-HBs antibodies, negative HBsAg levels and undetectable HBV viral load. Since treatment with plasmapheresis failed to induce remission, the patient was again treated with four weekly courses of rituximab. Due to several reports of HBV reactivation following rituximab therapy [11-13], prophylactic treatment with lamivudin was initiated. The patient did not need plasmapheresis after the second rituximab course and is now in complete remission for more than 19 months, with normal levels of liver enzymes.

METHODS

Levels of ADAMTS-13 activity were measured in plasma samples by chromogenic enzyme-linked immunosorbent

IVIG = intravenous immunoglobulin
HBV = hepatitis B virus

assay using TECHNOZYM® ADAMTS-13 Activity ELISA kit, Technoclone GmbH, Vienna, Austria. Antibodies against ADAMTS-13 were tested by TECHNOZYM® ADAMTS-13 INH ELISA kit (Technoclone GmbH, Vienna, Austria). Briefly, the detection wells were coated with recombinant ADAMTS-13. Antibodies against ADAMTS-13 from the plasma sample were recognized by conjugated anti-human IgG. The peroxidase level was determined by a chromogenic reaction that is proportional to the anti ADAMTS-13 antibody level.

DISCUSSION

Plasmapheresis, the standard treatment for TTP, improves prognosis and induces remission in more than 90% of patients. However, relapses are common [6]. There are no standard guidelines for the treatment and prevention of recurrent TTP. The options include plasmapheresis, corticosteroids alone or in combination with other immunosuppressive drugs (azathioprine, vincristine or cyclophosphamide), splenectomy or high-dose intravenous immune globulins [14-18]. However, an effective prophylactic regimen to prevent recurrent relapses is lacking.

More than 10 years ago an autoimmune process was implicated in the pathophysiology of some forms of TTP [2]. Arterial thrombi, the hallmark of TTP, consist of unusually large multimers of von Willebrand factor. These multimers are cleaved by a serum metalloprotease (ADAMTS-13) [1]. In patients with non-familial TTP, the presence of an autoantibody results in decreased levels of this metalloprotease [2]. Thus, targeting the activity of B lymphocytes which produce these autoantibodies may be beneficial in treating TTP. Rituximab, a chimeric antibody against the CD20 antigen presented on B lymphocytes, is used for the treatment of B cell lymphoma, idiopathic thrombocytopenic purpura, and other autoantibody-mediated autoimmune diseases [19-21]. Recent reports have described a beneficial response to rituximab with or without cyclophosphamide [22,23] in chronic-relapsing TTP. Rituximab attaches to the CD20 antigen in B lymphocytes, apparently triggering an immunological response, which eliminates B cells, including the clone that produces the anti-ADAMTS-13 antibodies.

All patients who fulfill the diagnosis of TTP are treated with plasmapheresis. According to the largest TTP-HUS registry from Oklahoma, the estimated risk for relapse is 41% at 7.5 years [24]. Most of the relapses occur during the first year after recovery. This group of relapsed TTP patients is a candidate for rituximab treatment, which in recent years has gained popularity.

This article describes four patients with recurrent or refractory TTP who achieved long-term remissions (13–72 months) following treatment with rituximab. Our experience with these patients is consistent with previously published data and substantiates a role for rituximab treatment in this potentially fatal disease, after relapses.

Patient 4 in our series had been previously exposed to hepatitis B virus. Reactivation of HBV leading to fulminant hepatic failure has been observed in these patients following treatment with rituximab [25]. In one series, 4.2% of 95 HBsAg-negative rituximab-treated lymphoma patients developed HBV reactivation [13]. Yeo et al. [11] reported that in HBsAg negative/anti-HBc positive lymphoma patients being treated with either CHOP or R-CHOP chemotherapy protocols, 5 of 21 patients treated with rituximab (R-CHOP) developed HBV reactivation (including one who died from hepatic failure), as compared to none of 25 patients receiving only CHOP [11]. Several reports have suggested that prophylactic treatment with lamivudin may reduce the rates of HBV reactivation [11,13]. In our patient 4, lamivudin was initiated at the time of the first rituximab cycle and continued for 6 months. HBV reactivation did not occur.

In conclusion, prolonged remissions of TTP-relapsing patients, following treatment with rituximab, as reported in this work, warrant further substantiation in future studies.

ELISA = enzyme-linked immunosorbent assay

Corresponding author:
Dr. A. Inbal
Thrombosis and Hemostasis Unit, Rabin Medical Center (Beilinson Campus), Petah Tikva 49101, Israel
Phone: (972-3) 937-7912
Fax: (972-3) 924-0145
email: aidainbal@hotmail.com

References

1. Furlan M, Robles R, Galbusera M, et al. von Willebrand factor-cleaving protease in thrombotic thrombocytopenic purpura and the hemolytic-uremic syndrome. *N Engl J Med* 1998; 339: 1578-84.
2. Furlan M, Robles R, Solenthaler M, Wassmer M, Sandoz P, Lammle B. Deficient activity of von Willebrand factor-cleaving protease in chronic relapsing thrombotic thrombocytopenic purpura. *Blood* 1997; 89: 3097-103.
3. Sadler JE. Von Willebrand factor, ADAMTS13, and thrombotic thrombocytopenic purpura. *Blood* 2008; 112: 11-18.
4. Cataland SR, Wu HM. Targeting the inhibitor of ADAMTS13 in thrombotic thrombocytopenic purpura. *Expert Opin Pharmacother* 2007; 8: 437-44.
5. Rock GA, Shumak KH, Buskard NA, et al. Comparison of plasma exchange with plasma infusion in the treatment of thrombotic thrombocytopenic purpura. Canadian Apheresis Study Group. *N Engl J Med* 1991; 325: 393-7.
6. Bell WR, Braine HG, Ness PM, Kickler TS. Improved survival in thrombotic thrombocytopenic purpura-hemolytic uremic syndrome. Clinical experience in 108 patients. *N Engl J Med* 1991; 325: 398-403.
7. Elliott MA, Heit JA, Pruthi RK, Gastineau DA, Winters JL, Hook CC. Rituximab for refractory and or relapsing thrombotic thrombocytopenic purpura related to immune-mediated severe ADAMTS13-deficiency: a report of four cases and a systematic review of the literature. *Eur J Haematol* 2009; 83: 365-72.
8. Ling HT, Field JJ, Blinder MA. Sustained response with rituximab in patients with thrombotic thrombocytopenic purpura: a report of 13 cases and review of the literature. *Am J Hematol* 2009; 84: 418-21.
9. Rabkin Y, Fradin Z, Zeidman A, Schwartz A, Cohen A, Mittelman M. Chronic relapsing thrombocytopenic purpura with severe neurological manifestations

and full recovery. *Int J Hematol* 2000; 71: 184-7.

10. Stein GY, Zeidman A, Fradin Z, Varon M, Cohen A, Mittelman M. Treatment of resistant thrombotic thrombocytopenic purpura with rituximab and cyclophosphamide. *Int J Hematol* 2004; 80: 94-6.
11. Yeo W, Chan TC, Leung NW, et al. Hepatitis B virus reactivation in lymphoma patients with prior resolved hepatitis B undergoing anticancer therapy with or without rituximab. *J Clin Oncol* 2009; 27: 605-11.
12. Colson P, Gerolami R, Moreau J, Borentain P, Brouqui P. Concurrent autochthonous acute hepatitis E and hepatitis B reverse seroconversion in an HIV-1-infected patient: one virus may hide another. *Int J Infect Dis* 2009; 14: e357.
13. Pei SN, Chen CH, Lee CM, et al. Reactivation of hepatitis B virus following rituximab-based regimens: a serious complication in both HBsAg-positive and HBsAg-negative patients. *Ann Hematol* 2009; 89: 255-62.
14. Dabrow MB, Wilkins JC. Hematologic emergencies. Management of hyperleukocytic syndrome, DIC, and thrombotic thrombocytopenic purpura. *Postgrad Med* 1993; 93: 193-4, 197-9, 202.
15. Moake JL, Rudy CK, Troll JH, et al. Therapy of chronic relapsing thrombotic thrombocytopenic purpura with prednisone and azathioprine. *Am J Hematol* 1985; 20: 73-9.
16. Thompson HW, McCarthy LJ. Thrombotic thrombocytopenic purpura. Potential benefit of splenectomy after plasma exchange. *Arch Intern Med* 1983; 143: 2117-19.
17. Van Gool S, Brock P, Van Laer P, Van Damme-Lombaerts R, Proesmans W, Casteels-Van Daele M. Successful treatment of recurrent thrombotic thrombocytopenic purpura with plasmapheresis and vincristine. *Eur J Pediatr* 1994; 153: 517-19.
18. Kolodziej M. Case report: high-dose intravenous immunoglobulin as therapy for thrombotic thrombocytopenic purpura. *Am J Med Sci* 1993; 305: 101-2.
19. Boumans MJ, Tak PP. Rituximab treatment in rheumatoid arthritis: how does it work? *Arthritis Res Ther* 2009; 11: 134.
20. Ahmed AR, Spigelman Z, Cavacini LA, Posner MR. Treatment of pemphigus vulgaris with rituximab and intravenous immune globulin. *N Engl J Med* 2006; 355: 1772-9.
21. Merrill JT, Neuwelt CM, Wallace DJ, et al. Efficacy and safety of rituximab in moderately-to-severely active systemic lupus erythematosus: the randomized, double-blind, phase ii/iii systemic lupus erythematosus evaluation of rituximab trial. *Arthritis Rheum* 2010; 62: 222-33.
22. Verbeke L, Delforge M, Dierickx D. Current insight into thrombotic thrombocytopenic purpura. *Blood Coagul Fibrinolysis* 2010; 21: 3-10.
23. Fakhouri F, Vernant JP, Veyradier A, et al. Efficiency of curative and prophylactic treatment with rituximab in ADAMTS13-deficient thrombotic thrombocytopenic purpura: a study of 11 cases. *Blood* 2005; 106: 1932-7.
24. George JN. How I treat patients with thrombotic thrombocytopenic purpura. *Blood* 2010; 116: 4060-9.
25. Sarrecchia C, Cappelli A, Aiello P. HBV reactivation with fatal fulminating hepatitis during rituximab treatment in a subject negative for HBsAg and positive for HBsAb and HBcAb. *J Infect Chemother* 2005; 11: 189-91.