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ORIGINAL ARTICLE

Integrated analysis of long-term safety in patients with chronic immune thrombocytopaenia (ITP) treated with the thrombopoietin (TPO) receptor agonist romiplostim

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Abstract A safety analysis of pooled data from clinical studies of romiplostim, a thrombopoietin (TPO) receptor agonist, in which patients with immune thrombocytopaenia (ITP) received romiplostim, placebo, or medical standard of care (SOC) Rodeghiero et al. (Eur J Haematol 91:423–436, 2013), has been updated. Included are data from 14 trials spanning 2002–2011; placebo- and SOC-arm data are pooled. Most patients ($n = 1059$) were female (61 %) and Caucasian (85 %); 38 % had undergone splenectomy; 23 were children. Mean (SD) baseline platelet count was 20.6 (16.5) $\times$ 10^9/L. Mean (SD) weekly dose of romiplostim was 4.2 (2.8) μg/kg; total exposure was 1520 patient-years. Overall, 921 patients received romiplostim only, 65 received placebo/SOC only, and 73 received placebo/SOC followed by romiplostim. Rates of haemorrhage (romiplostim, 205/100 patient-years; placebo/SOC, 263/100), thrombosis (both, 5.5/100 patient-years), haematological malignancy/myelodysplastic syndrome (romiplostim, 0.5/100 patient-years; placebo/SOC, 2.7/100), and non-haematological tumours (romiplostim, 2.2/100 patient-years; placebo/SOC, 3.6/100) were comparable among groups. Bone marrow reticulin was reported in 17 patients and collagen in one patient receiving romiplostim; one patient receiving placebo/SOC had reticulin reported. Three patients developed neutralizing antibodies to romiplostim, but not to endogenous TPO. This integrated analysis of the safety profile of romiplostim in patients with ITP is consistent with previously reported studies; no new safety concerns emerged.

Keywords Immune thrombocytopaenia (ITP) · Romiplostim · Thrombopoietin (TPO) · Platelet

Introduction

Immune thrombocytopaenia (ITP) is an autoimmune disorder characterised by low platelet counts due to increased platelet destruction and suboptimal platelet production,

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with clinical manifestations ranging from the absence of symptoms to serious internal bleeding [1]. Patients with ITP do not always respond to conventional treatments such as immunosuppressants (e.g. glucocorticoids), intravenous immunoglobulins (IVIg), and anti-Rh (D), each of which carries its own risk of side effects [1]. An alternative approach to treatment is the use of romiplostim, an Fc–peptide fusion protein (peptibody) that binds to and activates the thrombopoietin (TPO) receptor to stimulate platelet production [2]. Based on pivotal studies in splenectomised and non-splenectomised patients [3], romiplostim is approved in USA for the treatment of chronic ITP in adult patients who have had an insufficient response to corticosteroids, immunoglobulins, or splenectomy [4]. In the pivotal studies, treatment with romiplostim was associated with the overall response rates of 88 % in non-splenectomised patients [vs. 14 % for placebo (PBO)] and 79 % in splenectomised patients (vs. 0 % for PBO). As romiplostim can be used as a chronic treatment, long-term safety data are needed to guide clinical decision-making.

Results of a safety analysis of data pooled from 13 studies of romiplostim in ITP through June 2009 (718 patients, 653 of whom received romiplostim, representing 921.5 patient-years of exposure) were previously reported [5]. In that report, the rates of thrombosis and bone marrow reticulin did not appear to increase with long-term use of romiplostim, which is in keeping with the past analyses of thrombosis [6] and an ongoing study of bone marrow findings [7]. The current analysis comprises results from 14 studies through June 2011, involving 1059 patients with ITP; 994 received romiplostim, leading to a total exposure of 1520 patient-years. The resulting updated safety analysis of long-term treatment with romiplostim provides the most extensive clinical dataset to date of patients with ITP treated with a TPO receptor agonist.

Materials and methods

Patients and studies

Studies and methods were reported previously [5]. Briefly, data from 14 studies of romiplostim in ITP conducted between 1 July 2002 and 29 June 2011 were analysed. These studies were conducted in compliance with all regulatory obligations, including institutional review board and informed consent regulations at each investigational site. The parent studies included (with ClinicalTrials.gov identifiers) phase 2 dose-finding studies (NCT00111475 part B [8] and NCT00515203 [9]), phase 3 studies (NCT00102323 [3], NCT00102336 [3], NCT00603642 [10], and NCT00415532 [11]), and single-arm studies (NCT00111475 part A [8], NCT00117143 [12], NCT00305435 [13], NCT00508820 [14], and NCT00907478 [7]). Subsequent extension/follow-up studies included two open-label extension studies (NCT00116688 [15] and NCT00440037 [16]) and a bone marrow follow-up study (NCT00861224 [17] from the phase 3 studies NCT00102323 and NCT00102336 [3]). All but one study was conducted in adults [9].

Enrolment and exclusion criteria varied by study. Patients were diagnosed as having ITP per American Society of Haematology (ASH) guidelines [18] and were not receiving other investigational agents. Patients received weekly subcutaneous romiplostim or PBO along with concomitant or rescue medications as allowed per individual study protocols; standard of care (SOC) regimens varied. Platelet counts were targeted to be in the range of 50–200 $\times$ 10^9/L. In some earlier studies, the dose of romiplostim was adjusted from 1 to 15 μg/kg [3, 14, 15]; in all other studies, the dose of romiplostim was adjusted between 1 and 10 μg/kg. Patients received romiplostim on study for up to 283 weeks (5.4 years) and PBO/SOC for up to 18 months (1.5 years).

Assessments and statistical methods

Measurements of platelet counts and use of other ITP treatments were documented at each designated visit. Assessment of adverse events (AEs) was made when clinically apparent. Investigators evaluated AEs as to causality and severity (1 = mild to 5 = fatal). Specific AEs of interest included bone marrow reticulin and fibrosis (with bone marrow biopsies performed per investigators' discretion), haematological and non-haematological malignancies, myelodysplastic syndromes (MDS), thrombotic events (arterial and venous), and bleeding that was either grade 3 or greater or serious. A serious AE was defined as one that was fatal, life-threatening, required hospitalisation or prolonged a hospitalisation, or resulted in persistent or significant disability/incapacity, or other significant medical complication. Serious AEs were monitored by Amgen on an ongoing basis and all AEs were reviewed quarterly. Other safety data, including laboratory values, were reviewed for individual patients on an ad hoc basis and also on an ongoing basis for emerging trends and patient outliers. Results of an on-going study in which bone marrow biopsies were performed at baseline, and after 1, 2, or 3 years of treatment with romiplostim (i.e. three separate cohorts) as a primary endpoint will be reported separately once the final data are available; interim data from that study (i.e. not related to the bone marrow biopsies) are included in this analysis. Bone marrow findings per the modified Bauermeister grading scale from AE reports and available bone marrow biopsies from one of the open-label extension studies are reported. The

Table 1 Baseline characteristics

	Received romiplostim only ($N = 921$)	Received PBO/SOC only ($N = 65$)	Received PBO/SOC and then romiplostim ($N = 73$)	Total ($N = 1059$)
Age, years mean (SD)	52.0 (18)	54.4 (20)	50.1 (19)	52.0 (18)
Sex, female *n* (%)	561 (61)	38 (58.5)	49 (67)	648 (61)
Race, White/Caucasian *n* (%)	791 (86)	56 (86)	56 (77)	903 (85)
Baseline platelet count (10^9/L) mean (SD)	20.5 (17)	23.7 (16)	19.2 (11)	20.6 (16.5)
Years since ITP diagnosis, *n* (%)				
≤1 year	211 (23)	27 (42)	11 (15)	249 (24)
>1 year	710 (77)	38 (58)	62 (85)	810 (76)
Splenectomy	374 (41)	4 (6.2)	25 (34)	403 (38)

ITP immune thrombocytopaenia, *N* number of patients, *n* number of patients in a subset, *PBO* placebo, *SD* standard deviation, *SOC* standard of care

total number of bone marrow biopsies performed is not known because results were only reported if the outcome was considered to be an AE, with the exception of the extension study noted above.

Given the nature of this retrospective analysis, all analyses are descriptive, and only summary statistics are presented. Data from the PBO- and SOC-treatment arms are pooled. Unless otherwise indicated, results are adjusted for study duration and reported as events per 100 patient-years (calculated as the number of events/patient-year × 100) to reflect the unequal study duration between patients who received romiplostim and those who received PBO/SOC. When patients were enrolled in two consecutive studies, data from the parent and extension studies are combined. For patients who initially received PBO or SOC and then received romiplostim, data from the time before the patient received the first dose of romiplostim are included in the PBO/SOC group, and data beginning on the day of the first romiplostim dose onward are included in the romiplostim group, regardless of any subsequent change in treatment.

Results

Patients and disposition

The database included 1059 patients in total, of whom 921 received romiplostim only, 65 received PBO/SOC only, and 73 received PBO/SOC followed by romiplostim (i.e. in sequential studies). Thus, a total of 994 patients received romiplostim. For all 1059 patients, the mean (standard deviation [SD]) age was 52.0 (18.4) years; there were 23 paediatric patients (aged 0–17 years) and 294 geriatric patients (65 years or older) (Table 1). The mean (SD) baseline platelet count was 20.6 (16.5) × 10^9/L. The mean (SD) number of years since the diagnosis of ITP was 7.4 (9.3) years, with 76 % of the patients having had ITP for more than 1 year; 38 % of patients had a prior splenectomy. Prior treatments for ITP included glucocorticoids (97 %), IVIg (63 %), rituximab (26 %), danazol (22 %), anti-(Rh) D (20 %), azathioprine (20 %), and vincristine or vinblastine (11 %).

Of the 1059 patients (Fig. 1), 712 completed a parent study; the most common reasons for study discontinuation in romiplostim-treated patients were consent withdrawn ($n = 41$), AE ($n = 28$), and the need for alternative therapy ($n = 26$), while for PBO/SOC-treated patients, the most common reasons for study discontinuation were consent withdrawn ($n = 8$), death ($n = 7$), and AE ($n = 4$). Of those patients completing the parent study, 357 enrolled in an extension study.

Exposure

Patients received romiplostim for up to 283 weeks (5.4 years); mean (SD) treatment duration was 76 (64) weeks. The total exposure to romiplostim was 1520 patient-years. The mean (SD) dose of romiplostim used most frequently was 4.7 (3.6) μg/kg. The mean dose over time is shown in Fig. 2a and the dose used most frequently in Fig. 2b.

The mean (SD) treatment duration was 77 (73) weeks in patients who had splenectomy prior to the study ($N = 399$) and 75 (57) weeks in those who did not ($N = 595$). The mean (SD) doses of romiplostim used most frequently in the two groups were 5.0 (4.0) vs. 4.6 (3.3) μg/kg. The mean dose over time on study for the two populations is shown in Fig. 2c. The exposure in the two patient populations (splenectomised vs. non-splenectomised) appears to be comparable.

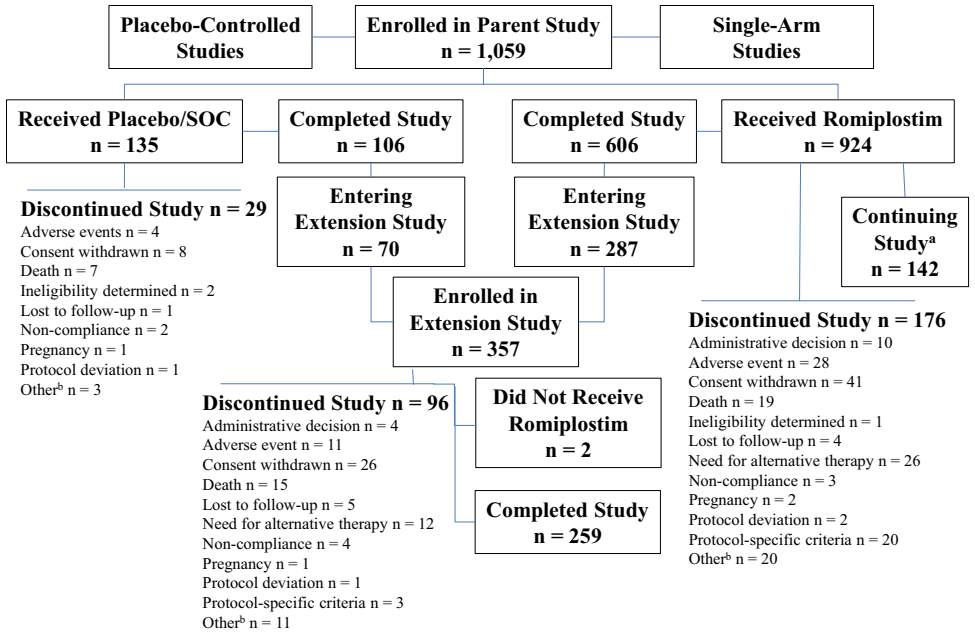

Fig. 1 Disposition. Patient disposition from parent studies through extension studies, when applicable, and reasons for discontinuation are shown. *n* number of patients

Overall AEs and deaths

The most frequent duration-adjusted AEs with romiplostim included headache, contusion, epistaxis, and nasopharyngitis (Supplementary Table 1). Of note, epistaxis occurred more frequently with PBO/SOC than romiplostim (duration-adjusted rates of 52.7 vs. 35.2 per 100 patient-years). Overall rates of serious AEs, treatment-related AEs, and treatment-related serious AEs were lower in patients who received romiplostim than in patients who received PBO/SOC (Table 2). Fatal AEs occurred in 40 patients receiving romiplostim and eight receiving PBO/SOC (Table 3). Five treatment-related fatalities were reported in patients receiving romiplostim (0.5 %) as opposed to none with PBO/SOC. Four of these five were described previously [5]. In brief, they were cases of (1) haemolysis in a 61-year-old splenectomised woman, (2) aplastic anaemia in a 75-year-old splenectomised woman, (3) myocardial infarction in a 60-year-old man, and (4) unstable angina in a 77-year-old woman. The fifth patient was a 79-year-old woman with a history of ITP for 6 years and chronic liver disease (hepatitis C virus serology positive), who developed intestinal ischaemia. Six months after her first dose of romiplostim, she presented to the emergency room in shock. She was found to have bilateral alveolar infiltrates. Despite treatment with antibiotics and supportive care, she died 1 day later. The investigator concluded that the shock, probably of respiratory origin, was not due to romiplostim, but that the pancreatic and intestinal ischaemia secondary to portal thrombosis might be related to romiplostim.

Haemorrhage and thrombosis

AEs classified as haemorrhage and thrombosis are described in Tables 2 and 4, and in Fig. 3. Five fatal bleeding events occurred during treatment with romiplostim; two events of intracranial haemorrhage, and one event each of cerebral haemorrhage, pulmonary haemorrhage, and decreased platelet count. One fatal cerebral haemorrhage was reported in a patient receiving PBO/SOC. The rates of haemorrhage decreased after the first 6 months with both romiplostim and PBO/SOC (Fig. 3a). The most common bleeding events in patients treated with romiplostim were contusion, epistaxis, and petechiae; the most frequent bleeding events in those receiving PBO/SOC were epistaxis, contusion, and petechiae.

The rate of thrombotic events was 5.5 per 100 patient-years, both in patients receiving romiplostim and those receiving PBO/SOC (Table 2, and shown as a function of time in Fig. 3b). The most common types of thrombotic events in patients receiving romiplostim were deep venous thrombosis, pulmonary embolism, and myocardial

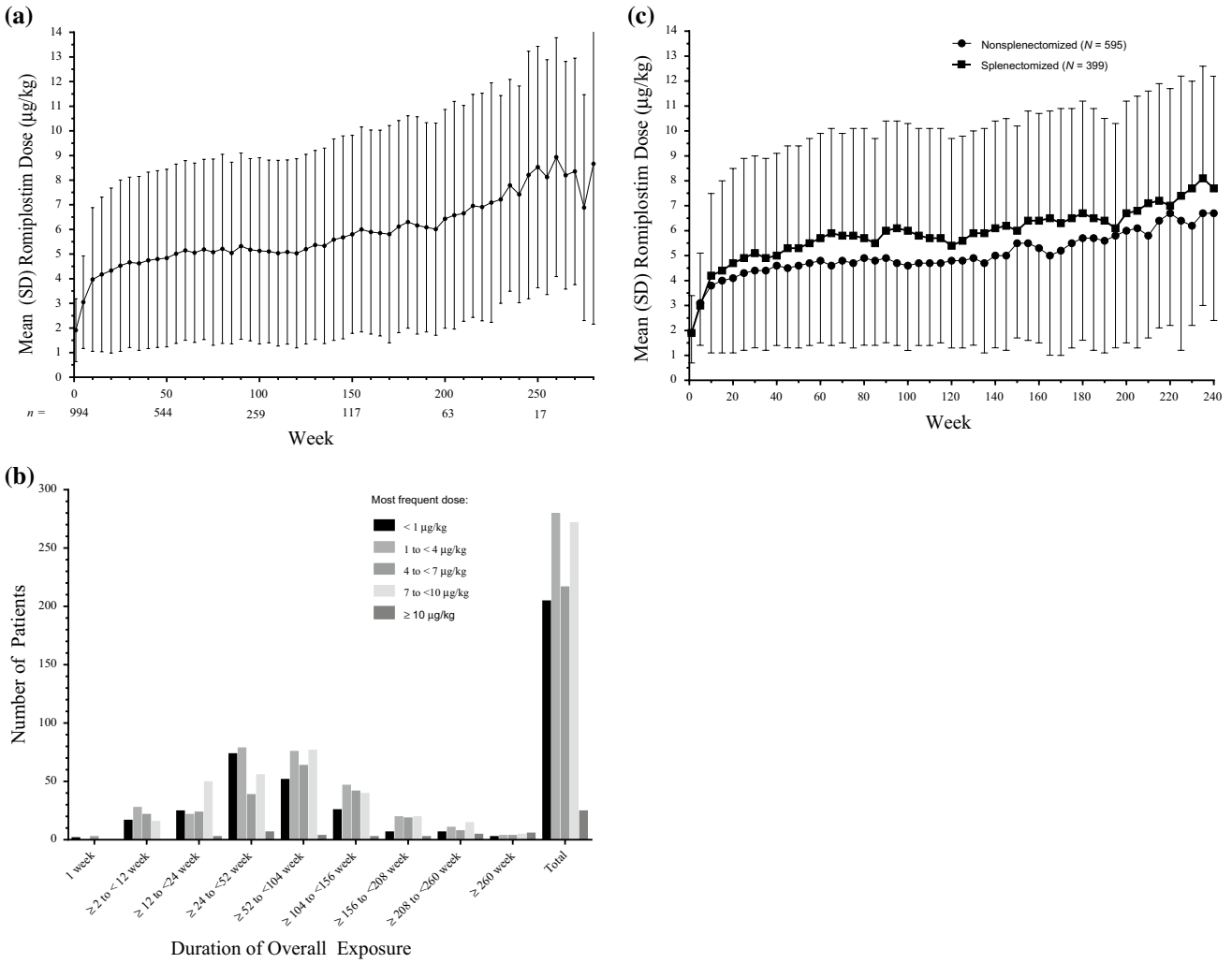

Fig. 2 Romiplostim dose. **a** Mean (SD) dose over time. **b** Most frequently used dose of romiplostim (µg/kg) over all treatment periods. If there was >1 most frequently used dose, the highest dose was chosen. Duration of overall exposure (weeks) was calculated as (last dose date − first dose date + 7)/7. If a patient was enrolled in multiple studies, the sum of treatment durations of the individual studies is shown. **c** Mean (SD) dose over time by splenectomy status. *n* number of patients, *SD* standard deviation, *Wk* week

infarction (full list in Table 4). Six thrombotic events occurred over the duration of the studies in patients receiving PBO/SOC, including venous thrombosis ($n = 2$), and deep vein thrombosis, pulmonary embolism, carotid artery occlusion, and cerebrovascular disorder (1 patient each) (Table 4). There were four events of portal vein thrombosis with romiplostim, two occurring in the first 6 months of treatment, one during months 7–12 of treatment, and the last in the third year of treatment; there were no reports of portal vein thrombosis in patients receiving PBO/SOC. The median (Quartile 1, Quartile 3) platelet counts most closely preceding the thrombotic events were 122.5 (56.0, 253.0) $\times$ 10^9/L in patients receiving romiplostim and 86.0 (11.0, 358.0) $\times$ 10^9/L in those receiving PBO/SOC. Thrombotic events occurred across a wide range of platelet counts in both romiplostim- and PBO/SOC-treated patients and did not appear to correlate with time above platelet thresholds (Supplementary Table 2). An exploratory analysis showed no difference in the rates of thrombotic events in patients who received a dose of romiplostim above 10 µg/kg at any time as compared with all those who received romiplostim [rates (95 % CI) of 3.6 (1.3–7.8) per 100 patient-years and 5.5 (4.4–6.8) per 100 patient-years, respectively].

Bone marrow events

Bone marrow results were collated from AE reports of abnormal bone marrow findings and from optional bone marrow biopsies from one of the open-label extension studies, in which both abnormal and normal bone marrow findings were reported. Post-baseline bone marrow

Table 2 Adverse events (AE)

	Romiplostim			PBO/SOC		
	$N = 994$; pt-yr = 1520			$N = 138$; pt-yr = 110		
	#	*r*	95 % CI	#	*r*	95 % CI
All AE	17,129	1127.1	1110–1144	1268	1152.3	1090–1218
Serious AE	910	60	56–64	107	97	80–118
Fatal AE	40	2.6	1.9–3.6	8	7.3	3.1–14.3
Treatment-related AE[a]	1739	114.4	109–120	168	152.7	131–178
Treatment-related serious AE[a]	118	7.8	6.4–9.3	18	16.4	9.7–25.9
Treatment-related fatal AEs[a]	5	0.3	0.1–0.8	0	0	0.0–2.7
AE leading to D/C IP (treatment-related)	83 (40)	5.5 (2.6)	4.4–6.8 (1.9–3.6)	8 (4)	7.3 (3.6)	3.1–14.3 (1.0–9.3)
AE leading to D/C study (treatment-related)	65 (35)	4.3 (2.3)	3.3–5.5 (1.6–3.2)	4 (0)	3.6 (0)	1.0–9.3 (0.0–2.7)
Haemorrhage						
Any grade	3115	205	198–212	289	263	233–295
Grade 3 or greater	182	12	10–14	19	17	10–27
Serious haemorrhage	151	9.9	8.4–11.7	15	13.6	7.6–22.5
Thrombotic/thromboembolic events	83	5.5	4.4–6.8	6	5.5	2.0–11.9
Serious thrombotic/thromboembolic events	61	4.0	3.1–5.2	2	1.8	0.2–6.6
Bone marrow reticulin/collagen[b]	18	1.3	0.8–2.1	1	0.9	0.0–5.1
Non-haematological tumours	34	2.2	1.6–3.1	4	3.6	1.0–9.3
Haematological malignancies/MDS	7[c]	0.5	0.2–1.0	3[d]	2.7	0.6–8.0
Cataracts	34	2.2	1.6–3.1	1	0.9	0.0–5.1

AE adverse event, *CI* confidence interval, *D/C* discontinuation, *IP* investigational product, *MDS* myelodysplastic syndrome, *N* number of patients, *#* number of events, *PBO* placebo, *Pt-yr* total patient-years on study, *r* rate per 100 patient-years, *SOC* standard of care

[a] Attribution of relatedness was at the discretion of the treating physician

[b] Bone marrow biopsies were performed per investigators' discretion. Analysis excludes study NCT00907478, in which bone marrow biopsies were collected both at baseline and after 1, 2, or 3 years of romiplostim (i.e. three different cohorts). Bone marrow biopsies in other studies were collected at the investigator's discretion. Romiplostim, $N = 825$, pt-yr = 1388; PBO/SOC, $N = 138$, pt-yr = 110

[c] Includes two events of multiple myeloma and one event each of lymphoma, acute myeloid leukaemia, B cell lymphoma, chronic lymphocytic leukaemia, and non-Hodgkin's lymphoma

[d] Includes one event each of multiple myeloma, lymphoma, and MDS

biopsies were collected from 48 individuals. Seventeen events of reticulin and one of collagen were reported in patients receiving romiplostim, and one case of reticulin was reported in a patient receiving PBO (Table 2; Fig. 3b). Bone marrow finding event-rates for those receiving romiplostim ($N = 825$) and a subgroup of patients with a maximum romiplostim dose over 10 μg/kg ($N = 61$) were 1.3 (18 cases) per 100 patient-years (95 % CI 0.8–2.1) and 3.6 (6 cases) per 100 patient-years (95 % CI 1.3–7.8), respectively.

Fifteen of the 17 cases of reticulin and one case of collagen in patients receiving romiplostim were reported as AEs; two additional cases of reticulin were identified among reports of optional bone marrow biopsies in the open-label extension study. The bone marrow biopsy reports also revealed that one patient who had been receiving PBO in a pivotal trial subsequently had reticulin reported in a bone marrow biopsy performed before receiving romiplostim. A biopsy performed in this same patient after 3 months of romiplostim showed no change in reticulin.

All AEs involving reticulin in patients receiving romiplostim were considered treatment-related; of the 10 that were deemed serious (i.e. fatal, life-threatening, or requiring hospitalisation), four patients were hospitalised: (1) one with gingival bleeding treated with platelets and methylprednisolone, (2) one with bleeding from the ear treated with platelets, IVIg, and corticosteroids, (3) one with immature cells (i.e. myeloblasts 1 %, promyelocytes 1 %, myelocytes 2 %, metamyelocytes 6 %, bands 4 %) treated with prednisone, and (4) one with a platelet count of 2×10^9/L treated with platelet transfusion. In one patient, alternative treatment for ITP (corticosteroids and IVIg) was instituted on an outpatient basis. In the remaining five patients, either drug was discontinued (four patients) or no action was taken (one patient). Romiplostim was discontinued in nine of the 15 patients with reticulin AEs, the dose was reduced in two patients, and the remaining four

Table 3 Fatal adverse events (AEs)

n (*r*)	Romiplostim $N = 994$; pt-yr = 1520	PBO/SOC $N = 138$; pt-yr = 110
Cardiac		
Cardiac arrest	1 (0.1)	–
Cardiac failure	1 (0.1)	–
Cardiac tamponade	1 (0.1)	–
Cardio-respiratory arrest	–	1 (0.9)
Congestive cardiac failure	2 (0.1)	–
Left ventricular failure	–	1 (0.9)
Myocardial infarction	3 (0.2)	–
Sudden death	1 (0.1)	–
Unstable angina	1 (0.1)	–
Haemorrhage		
Cerebral haemorrhage[a]	1 (0.1)	1 (0.9)
Decreased platelet count[a]	1 (0.1)	–
Intracranial haemorrhage[a]	2 (0.1)	–
Pulmonary haemorrhage[a]	1 (0.1)	–
Infection		
Cytomegalovirus infection	1 (0.1)	–
Fungal sepsis	2 (0.1)	–
Meningitis listeria	1 (0.1)	–
Osteomyelitis	1 (0.1)	–
PML	1 (0.1)	–
Pneumococcal sepsis	1 (0.1)	–
Pneumonia	2 (0.1)	–
Pneumonia streptococcal	1 (0.1)	–
Primary atypical pneumonia	–	1 (0.9)
Neoplasm		
Malignant hepatic neoplasm	1 (0.1)	1 (0.9)
Malignant lung neoplasm	1 (0.1)	–
Metastatic lung cancer	–	1 (0.9)
Renal		
Renal failure	3 (0.2)	–
Renal failure acute	1 (0.1)	–
Thrombotic		
Intestinal infarction	1 (0.1)	–
Intestinal ischaemia	1 (0.1)	–
Intracranial venous sinus thrombosis	1 (0.1)	–
Ischaemic stroke	1 (0.1)	–
Pulmonary embolism	–	1 (0.9)
Other		
Acute respiratory distress syndrome	1 (0.1)	–
Aplastic anaemia	1 (0.1)	–
Brain oedema	1 (0.1)	–
Death	1 (0.1)	–
Haemolysis	1 (0.1)	–
Hepatic failure	–	1 (0.9)

AEs were coded using MedDRA version 14.0. Only AEs starting after the first dose of the investigational product were tabulated

ICH intracranial haemorrhage, *n* number of patients, *PBO* placebo, *PML* progressive multifocal leukoencephalopathy, *pt-yr* total patient-years on study, *r* rate per 100 patient-years, *SOC* standard of care

[a] The two platelet counts prior to these events were 3×10^9/L and 3×10^9/L for the first ICH, 5×10^9/L and 7×10^9/L for the second ICH, 5×10^9/L and 8×10^9/L for the cerebral haemorrhage, 110×10^9/L and 60×10^9/L for the pulmonary haemorrhage, and 19×10^9/L and 17×10^9/L for the death due to decreased platelet count. For the fatal cerebral haemorrhage in PBO/SOC, the two platelet counts prior to this event were 15×10^9/L and 9×10^9/L

Table 4 Thrombotic/thromboembolic events (T/TE)

n (*r*)	Romiplostim $N = 994$; pt-yr = 1520	PBO/SOC $N = 138$; pt-yr = 110
Total # T/TE reported	83 (5.5)	6 (5.5)
T/TE #/rate by platelet count ($\times 10^9$/L) closest to T/TE within the previous 3 months[a]		
<20	11 (0.7)	2 (1.8)
<30	12 (0.8)	2 (1.8)
<50	17 (1.1)	3 (2.7)
50–100	11 (0.7)	1 (0.9)
100–200	23 (1.5)	–
200–400	23 (1.5)	1 (0.9)
>400	9 (0.6)	1 (0.9)
T/TE by type		
Arterial		
Myocardial infarction	9 (0.6)	–
Transient ischaemic attack	5 (0.3)	–
Acute myocardial infarction	4 (0.3)	–
Coronary artery occlusion	1 (0.1)	–
Peripheral embolism	1 (0.1)	–
Carotid artery occlusion	–	1 (0.9)
Ischaemic stroke	1 (0.1)	–
Venous		
Deep vein thrombosis	17 (1.1)	1 (0.9)
Pulmonary embolism	13 (0.9)	1 (0.9)
Thrombophlebitis	5 (0.3)	–
Portal vein thrombosis	4 (0.3)	–
Transverse sinus thrombosis	1 (0.1)	–
Venous thrombosis	1 (0.1)	2 (1.8)
Thrombophlebitis superficial	4 (0.3)	–
Pulmonary thrombosis	2 (0.1)	–
Intracranial venous sinus thrombosis	1 (0.1)	–
Venous thrombosis limb	1 (0.1)	–
Mixed arterial/venous		
Cerebrovascular accident	6 (0.4)	–
Hemiparesis	2 (0.1)	–
Thrombosis	2 (0.1)	–
Intestinal infarction	1 (0.1)	–
Cerebrovascular disorder	–	1 (0.9)
Splenic infarction	1 (0.1)	–
Thrombosis in device	1 (0.1)	–

n number of events, *N* total number of patients, *Pt-yr* total patient-years on study, *r* rate per 100 patient-years, *PBO* placebo, *SOC* standard of care

[a] Duration of time that patients spent in each platelet count category could not be assessed as platelet counts were not performed frequently enough to allow such analysis

patients continued taking romiplostim as before. Of the nine patients who discontinued romiplostim, six of those also discontinued the study at that time, so there was no follow-up. For the remaining three patients who discontinued romiplostim and stayed on study, one patient was reported as doing well 4 months later but did not have a follow-up bone marrow biopsy, a second patient had no change in a biopsy performed 2 months later and was reported as doing well 1½ years later, and for the third patient, there was no additional information. Also, there was no additional information for the two patients who had a decrease in dose.

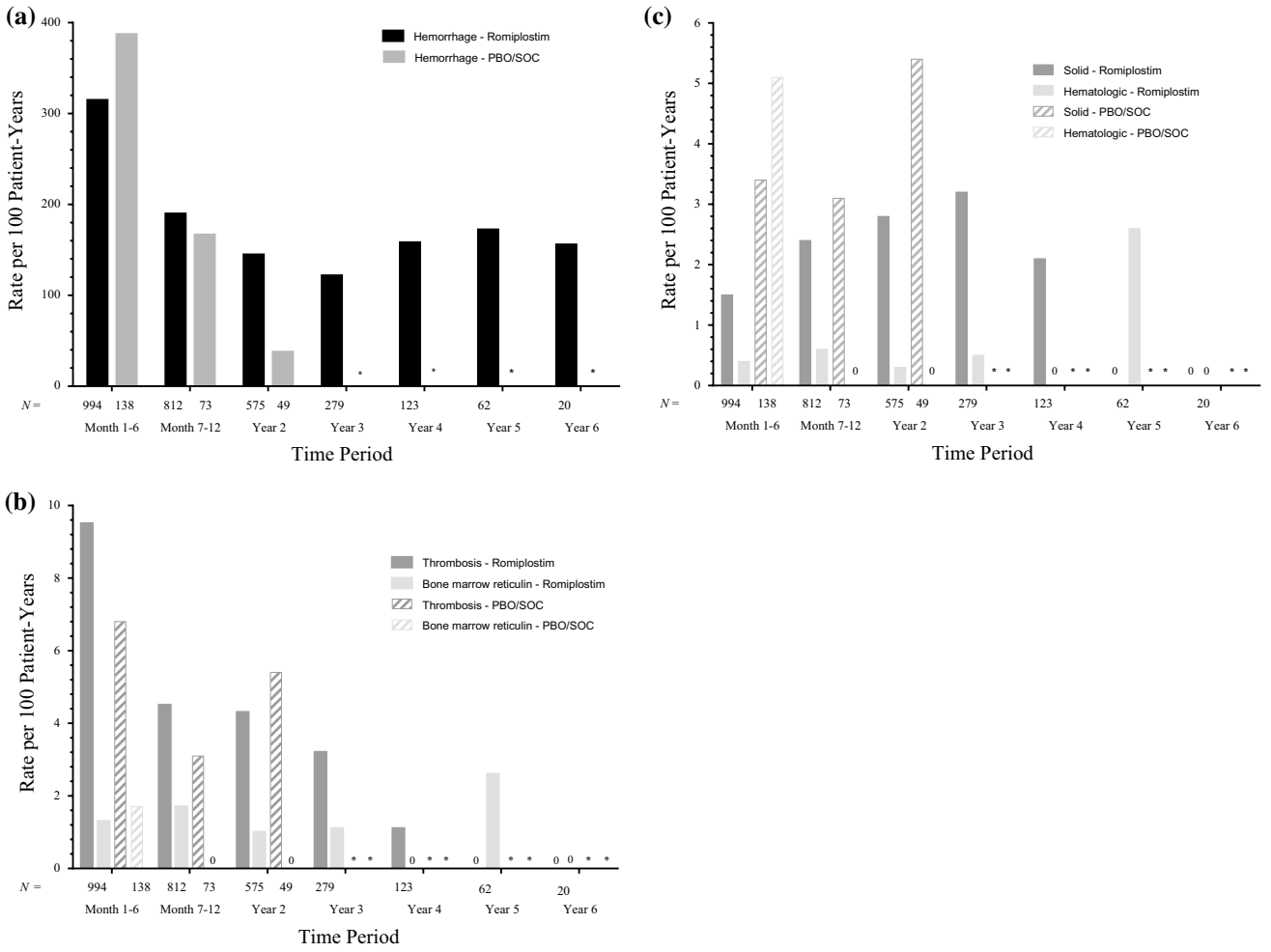

Fig. 3 Adverse events over time. **a** Rate of bleeding (any grade) over time. **b** Rate of thrombosis and reticulin over time. Ns under the graph refer to the total number of patients at that time—the actual number of patients in the bone marrow analysis was fewer at early time points, as the bone marrow study was excluded (months 1–6, $N = 825$; months 7–12, $N = 667$; year 2, $N = 528$; the remainder were the same). **c** Rate of malignancies over time. *Asterisk* no patients in that group. *PBO* placebo, *SOC* standard of care

Grading of reticulin was reported in ten patients, often provided as a range; with highest grades of 4 (in one patient), 3 (in four patients), 2 (in two patients), and 1 (in three patients). Each of the 15 patients with reticulin events had received romiplostim in maximum doses ranging from 5 to 18 μg/kg, including four patients whose doses exceeded current recommendations (i.e. >10 μg/kg) [4]. For the additional two cases of reticulin that were not identified by investigators as AEs, one patient received romiplostim in doses ranging from 0.2 to 15 μg/kg over the course of 184 weeks at the time of the reticulin finding and the second received romiplostim at a dose of 3 μg/kg over 72 weeks; after the biopsies were performed, the patients continued to receive romiplostim. Data regarding the interval between initiation of treatment and detection of reticulin or collagen (one case) were available for all 18 cases. This interval ranged from 29 to 2066 days. Two cases of reticulin were reported within the first month of treatment, one in months 2 and 3, none in months 4–6, five in month 7–1 year, and ten after a year. Detailed data from 12 of the reticulin AEs and the single event of bone marrow collagen, which occurred after the patient received romiplostim for 9 months at doses of up to 11 μg/kg, have been described previously [5].

In the open-label extension study in which optional bone marrow biopsies were collected from 40 patients, there were 29 patients who had bone marrow biopsies but no findings of reticulin that were either reported as an AE or noted in the accompanying pathology reports. Demographic characteristics of these 29 patients were compared with those of 18 patients treated with romiplostim with findings (17 with reticulin and one with collagen) derived from all studies (i.e. not just the 11 patients from the open-label extension, as reported above and in Table 2). There

were no significant differences in mean age, baseline platelet count, and duration of ITP (data not shown). Major limitations of this comparison include that the two groups were not randomised and that no bone marrow biopsies had been performed just prior to treatment in the 18 romiplostim-treated patients with findings of reticulin/collagen.

Rates for haematological malignancies were 0.5 per 100 patient-years (seven events) for patients receiving romiplostim and 2.7 per 100 patient-years (three events) for those receiving PBO/SOC (Fig. 3c; Supplementary Table 3). One event of multiple myeloma was considered to be related to romiplostim.

Miscellaneous AEs

None of the 34 events of non-haematologic tumours in 22 romiplostim-treated patients were considered to be related to romiplostim (Fig. 3c; Supplementary Table 3). Thirty-four events of cataracts were reported with romiplostim, two of which were considered serious; there was one case with PBO/SOC.

Three patients developed neutralizing antibodies to romiplostim but not endogenous TPO (Table 2; Supplementary Fig. 1). The one patient who continued romiplostim continued to respond, a second patient was at the end of study when the antibodies were detected, and the third maintained platelet counts even though romiplostim had been discontinued.

Discussion

This integrated analysis provides the results of all completed clinical studies performed to date in which patients with ITP received romiplostim. Updated safety data are provided for over 1000 patients with a mean time on study of 76 weeks. The results reported here are similar to those observed in individual studies and in a prior pooled analysis [3, 5, 7–17]. There was no increase in the time-adjusted rates of AEs previously designated to be of interest, including bone marrow reticulin, thrombosis, and malignancy, with longer term treatment with romiplostim (up to more than 5 years for some patients), and no new safety signals emerged. While the mean dose appeared to increase over time, this may reflect a higher dose employed in those patients who had been on romiplostim the longest. Those patients had entered earlier trials in which higher doses were allowed. Further, those patients may have had more refractory disease, particularly as some of the later trials required that patients not have had splenectomies. This increase in dosage was also noted when 292 patients in one of the open-label extension studies were analysed based on time of study entry [15].

That treatment with romiplostim did not increase the rates of thrombosis is important in view of the emerging concept that ITP is itself a prothrombotic state. In a recent meta-analysis, patients with ITP not receiving TPO receptor agonists showed an approximate two-fold increased risk of venous thromboembolism and 50 % increased risk of arterial thromboembolism as compared with the general population ([19], source studies [20–24]). The most common thrombotic events that were observed in patients receiving romiplostim reflect those most commonly reported in the general population, i.e. myocardial infarction and transient ischaemic attack on the arterial side and deep vein thrombosis and pulmonary embolism on the venous side, with the one exception being the four cases of portal vein thrombosis in patients receiving romiplostim. In some of these cases, there was an interval of several weeks between the last recorded platelet count and the thrombotic event, making it difficult to determine if there was a relationship between the platelet count and the thrombotic event. However, the available data show no general predilection for thrombosis to occur at higher platelet counts. Thrombotic events did occur more frequently early during treatment when increases in platelet count first occurred, and the rise may have been more rapid. This suggests the possibility that some patients have a pre-existing vulnerability to thrombosis such that when platelet count increases with any treatment, such as with romiplostim in these studies, thrombotic events are more likely to occur.

Although a number of abnormal bone marrow findings were reported in patients receiving romiplostim (17 reticulin and one collagen out of 48 patients biopsied), it is hard to interpret the significance of this observation. Only a limited number of biopsy results were available from either AE reports or the optional biopsies performed in one study (biopsies were obtained in 48 of the 994 romiplostim-treated patients), many biopsies were performed as a follow-up to clinical developments at the discretion of the principal investigator rather than systematically, and baseline pre-treatment biopsies for comparison were generally not available. Some cases of reticulin were designated grades 1–2, which is found commonly in patients with ITP who have not received treatment with TPO receptor agonists [25]. Whether reticulin is detected more frequently in patients receiving higher doses or more protracted courses of romiplostim cannot be assessed from this study. Six out of 18 patients in whom bone marrow reticulin/collagen was reported had received a maximum dose of romiplostim above 10 μg/kg. These patients, who required doses above current recommendations, could have had pre-existing bone marrow pathology affecting their responsiveness to romiplostim. However, the low number of patients who received

doses of romiplostim over 10 μg/kg ($n = 61$) makes interpretation difficult. It is anticipated that results from the 3-year prospective study of romiplostim's effects on the bone marrow will provide more information on incidence and risk factors for the development of reticulin fibrosis.

The issue of whether reticulin and collagen fibrosis are more likely to develop in those patients who are successfully treated with TPO receptor agonists rather than other forms of treatment is unsettled. Recently, Ghanima et al. reported an association between treatment with TPO receptor agonists and grade 2/3 myelofibrosis as evaluated using the European Consensus Scale in a single-centre study of 66 patients with ITP with no associated clinical findings [26]. Of note, few cases of collagen were identified (five of 245 samples). Thus, most of the 34 biopsies classified as myelofibrosis (MF)-2 or MF-3 were positive for reticulin only. Results from prospective, systematic evaluations of bone marrow showed few notable changes after treatment with romiplostim [7] or eltrombopag [27] (also a TPO receptor agonist). As additional results become available, such as 3-year data from the romiplostim trial and 2-year data from the eltrombopag trial, a more complete picture of the effects of stimulating the TPO receptor on bone marrow reticulin will emerge.

There are several limitations of our analysis. First, most of these studies were not designed to systematically collect data relating to specific AEs of interest. Second, as the average follow-up time per patient was much longer in the romiplostim group than in the PBO/SOC group, it is difficult to make meaningful comparisons for rare events or events with delayed onset. Lastly, there were far more romiplostim-treated patients than PBO/SOC-treated patients. Therefore, as would be expected, patients receiving romiplostim experienced a wider variety of AEs than those receiving PBO/SOC, likely both due to the increased number of patients and the greater time of exposure and follow-up.

In summary, this analysis supports previously reported findings regarding the incidence of thrombosis and bone marrow reticulin in patients receiving romiplostim for up to 5 years. Results from ongoing prospective studies of bone marrow biopsies will provide more information as to the incidence of reticulin, and other studies will provide insight into the frequency with which treatment with romiplostim can be discontinued while maintaining platelet counts [28].

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Compliance with ethical standards

Conflict of interest This study was funded by Amgen. Douglas B. Cines has been a consultant for Amgen, GlaxoSmithKline, Symphogen, Cangene, Rigel, and Eisai. Terry Gernsheimer is a consultant for Amgen, Symphogen, Clinical Options, GlaxoSmithKline, and Cangene; received honoraria from Amgen, Laboratorios Raffo SA, and Hemedicus Corporation; and received research funding from Shionogi. Jeffrey Wasser has served on the advisory board and speaker's bureau for Amgen and is also a consultant for Amgen. Bertrand Godeau is a consultant for Amgen, GlaxoSmithKline, LFB, and Roche and has received research funding from Roche. Drew Provan has honoraria from GlaxoSmithKline and Amgen and is a stockholder of GlaxoSmithKline. Roger Lyons is a consultant for Amgen. Ivy Altomare has served on the advisory board and speaker's bureau for Amgen. Xuena Wang is an employee of and shareholder in Amgen; Angela Lopez was an employee of and is a shareholder in Amgen.

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