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Avatrombopag for the salvage treatment of platelet transfusion refractoriness

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Abstract

Background: Platelet transfusion refractoriness (PTR) is a life-threatening and intractable condition in hematological patients. Thrombopoietin receptor agonists such as avatrombopag promote platelet production and modulate immune intolerance. However, its application in PTR has not been extensively studied.

Objectives: We aimed to compare the platelet response (PR) as well as bleeding events and mortality rate between the best available therapies (BATs) and avatrombopag (Ava) treatments in refractory PTR patients.

Design: A total of 71 refractory PTR patients were enrolled at Nanfang Hospital. Intravenous immunoglobulin, steroids, and human leucocyte antigen-matched platelet transfusions were administered to 30 patients in the BATs group. The Ava group included 41 patients.

Methods: Data of refractory PTR patients were retrospectively collected. The primary endpoint was PR (defined as an increase of platelet count to $\geqslant 50 \times 10^9$/L without platelet transfusion support for 7 consecutive days). Secondary endpoints included platelet-transfusion independence rate, cumulative platelet transfusion units, World Health Organization bleeding grades, adverse events, overall survival (OS), and bleeding event-free survival (EFS).

Results: There were 75.6% and 13.3% refractory PTR patients who reached PR within 3 months in Ava and BATs groups. The median platelet counts were significantly higher in Ava group from day 7. Platelet-transfusion independence rate in Ava was higher than BATs group. The median cumulative platelet transfusion unit in Ava was lower than that of BATs group. The OS and bleeding events-free EFS rate of Ava group improved within 3 months as compared to BATs group. Cox proportional hazards regression analysis revealed that Ava therapy was a protective factor for the OS and EFS. No primary disease progression or termination of avatrombopag was observed due to intolerability.

Conclusion: Our study suggests that avatrombopag is an effective and safe treatment option for refractory PTR patients.

Plain language summary

Avatrombopag in platelet transfusion refractoriness

PTR is a challenging clinical issue in patients with hematologic disorders which increases early death and hospitalization costs. Thrombopoietin receptor agonists have shown inspiring effects in treating thrombocytopenia. However, there are few studies focused on the application of these drugs in PTR patients. In this study, we investigated 71 patients with PTR in which 30 patients received the best available therapies, while 41 patients received avatrombopag treatment. We found that avatrombopag increases platelet response rate, reduces platelet transfusions dependence and occurrence of severe bleeding events, as well as improves overall survival rate and event free survival in PTR patients. Avatrombopag also exhibited good tolerance and safety. We reported for the first time that avatrombopag was an effective and safe treatment in PTR, which may also help to expand the clinical application of TPO-RAs.

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Background

Platelet transfusion refractoriness (PTRs) is a severe complication in patients with malignant disease, severe infection, and autoimmune disease.[1,2] The trial to reduce alloimmunization to platelets study group defined PTR as a corrected count increment (CCI) of less than 5×10^9/L 1 h after two consecutive transfusions of the same fresh platelets with ABO.[3] The incidence of PTR is reported to vary from 4.8% to 54.7%.[4–6] PTR is also associated with serious adverse events, including severe bleeding, relapse, or deterioration of malignant disease due to reduced, delayed, or discontinued chemotherapy as well as an increased risk of early death. In addition, it may lead to extended hospital stays and increased medical expenses.[7–9]

Currently, the most effective treatment of PTR is human leucocyte antigen (HLA)-matched platelet transfusions. However, the efficacy of the therapy is sporadic and difficult to achieve.[10,11] Other strategies include massive transfusions combined with intravenous immunoglobulins (IVIGs), steroids, plasma exchange, rituximab, recombinant human thrombopoietin, or bortezomib. Nevertheless, owing to the complicated mechanisms involved, the efficiency of these strategies remains largely unsatisfactory.[10,12–15] Both immunological and non-immune processes contribute to PTR. Of note, non-immune causes include sepsis, diffuse intravascular coagulation, medication, and splenomegaly.[2,10] Most of these non-immune causes may be improved by treating the underlying cause. Still, the associated immune mechanisms are relatively ambiguous. Recent studies suggest that the immune factors may include Fc receptor (FcR)-mediated enhancement of phagocytosis and T cell-mediated immune imbalance, which may further lead to megakaryopoiesis and thrombopoiesis destruction.[16–18] FcR-mediated enhancement of phagocytosis-induced PTR can be treated with IVIG and steroids[19,20]; yet, no recognized treatment scheme for PTR mediated by T cell dysregulation is currently available.

Thrombopoietin-receptor agonist (TPO-RA) mimics the endogenous thrombopoietin by stimulating the proliferation and differentiation of megakaryocytes, thereby increasing platelet production.[21,22] It has been widely used to treat thrombocytopenia in various diseases, including immune thrombocytopenia, chronic liver disease, and severe aplastic anemia.[23,24] In addition to the direct stimulation of megakaryopoiesis, TPO-RAs also have immune regulatory functions, including T-cell anergy,[25,26] platelet (microparticles)-mediated immunoregulation,[27] release of platelet transforming growth factor β (TGF-β),[28] induction and improved immunosuppressive function of adaptive Tregs,[29] and increased regulatory B-cell function.[29] Avatrombopag (Ava), a type of TPO-RA, is superior to other TPO-RAs for its reduced hepatotoxicity and nephrotoxicity, minimum drug–drug interaction, and dietary restriction.[9] Therefore, it may be a better choice for hematological patients with polypharmacy. Notably, a multicenter open-label study has confirmed that Ava is effective and tolerable as a treatment for chemotherapy-induced thrombocytopenia.[30] However, the effects of avatrombopag in PTR have not been reported yet. Therefore, we conducted a retrospective study to compare the platelet response (PR) and mortality rate between the use of avatrombopag along with the best available therapies (BATs) for refractory PTR patients.

Methods

Study design and patients

The Ethics Committee of the Nanfang Hospital, affiliated with Southern Medical University, approved the study. The requirement to obtain informed consent was waived due to the retrospective nature of the study and was deemed exempt from review by the Ethics Committee of the Nanfang Hospital of Southern Medical University. In total, 71 patients with refractory PTR from April 2017 to September 2022 were enrolled at our institution. IVIG, steroids, and HLA-matched platelet transfusions were administered to 30 patients in the BATs group. The Ava group included 41 patients who received avatrombopag treatment.

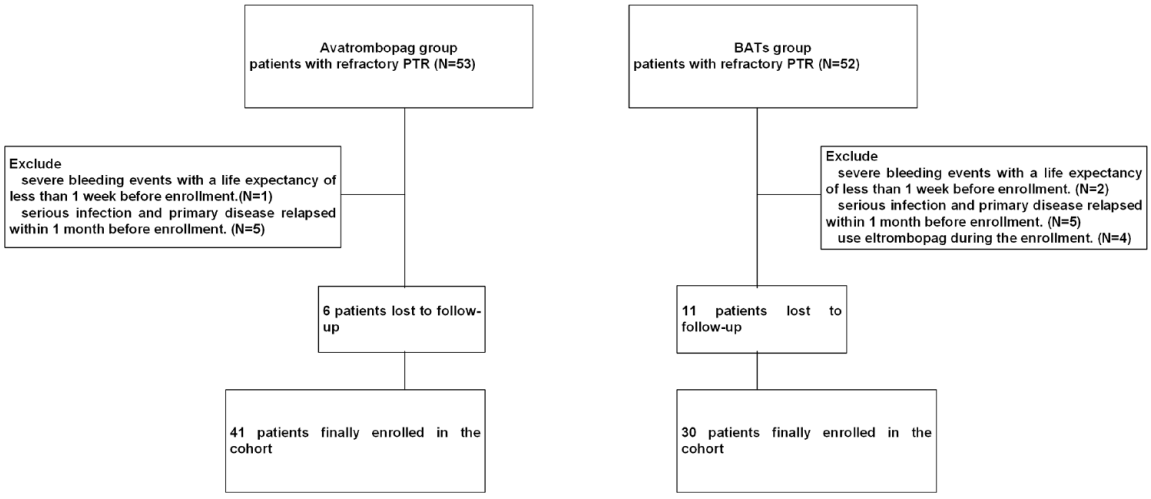

Figure 1. Flow chart of patient inclusion. Exclusion process for two groups of patients.

The inclusion criteria were as follows[9]: (1) age $\geqslant$18 years; (2) diagnosis of refractory PTR (defined here as a 1 h CCI $<5\times10^9$/L after transfusion of two consecutive rounds of HLA-matched platelets). Exclusion criteria were as follows[9]: (1) cardiovascular disease (including acute coronary syndrome, serious cardiac arrhythmia, acute myocarditis), thromboembolic disease (including arterial thrombotic, venous thrombotic, and thrombotic microangiopathy), or any other illness requiring systemic anticoagulation within 6 months; (2) serious infection, disseminated intravascular coagulation, or relapse and progression of primary disease with a life expectancy of less than 1 week; (2) a history of lupus anticoagulant or antiphospholipid syndrome; (3) liver dysfunction (total bilirubin, aspartate aminotransferase, or alanine aminotransferase three times over the upper limit of normal), as well as renal dysfunction (creatinine 1.5 times the upper limit of normal). The inclusion procedure is shown in Figure 1.

Avatrombopag and BATs treatment

Avatrombopag therapy was commenced at an oral dose of 20–40 mg once daily, which was subsequently increased or decreased in accordance with the response and tolerance of patients, with a maximum daily dose of 60 mg. When platelet counts exceeded 100×10^9/L, or adverse events were associated with the treatment, and the drug was tapered or immediately discontinued. Cross-matching platelet transfusion was allowed for patients with platelet counts of $<20\times10^9$/L or patients with clinical evidence of bleeding.

Patients in BATs groups were treated by the following therapies, alone or in combination: (1) IVIG was administered at a dose of 0.4 g/kg/day for 3–5 days, (2) prednisone (1–2 mg/kg/day) or equivalent dose of methylprednisolone, and (3) HLA-match or cross-matching platelet transfusion. In both groups, transfusion of red blood cells was allowed for patients with hemoglobin levels of $\leqslant$60 g/L.

Endpoints

The primary endpoint was PR, also known as treatment response. It was defined as an increase of platelet count to $\geqslant50\times10^9$/L without platelet transfusion support for 7 consecutive days after enrollment. Secondary endpoints included platelet count on days 7, 14, 21, and 28, respectively, platelet-transfusion independence rate, cumulative platelet transfusion units, World Health Organization (WHO) bleeding grades, adverse events, overall survival (OS), and bleeding event-free survival (EFS). OS is defined as the time from the initiation of treatment to death from any cause or to the termination of follow-up. EFS is defined as the period from the initiation of treatment to either death for any reason, the end of follow-up, or appearance of WHO grade 3–4 bleeding events. The criteria for platelet-transfusion independence was defined as follows: a consecutive 3-day period with a platelet count $\geqslant20\times10^9$/L without the need for platelet transfusion and the absence of bleeding tendencies. Platelet transfusion was measured in units, where each unit contained 2.5×10^{10} platelets. As a

criterion for the efficacy of platelet transfusion, CCI was calculated using a recognized calculation method.[3]

The WHO scale was used to grade bleeding. Petechiae was rated as grade 1 on this scale, minor blood loss as grade 2, gross blood loss as grade 3, debilitating blood loss as grade 4, and severe cerebral bleeding as grade 5.[31] Adverse events were monitored and assessed according to the Common Terminology Criteria for Adverse Events version 5.0 (CTCAE5.0).[32]

Safety and tolerability

We assessed disease status of acute myelocytic leukemia (AML), acute lymphoblastic leukemia, and myelodysplastic syndrome (MDS) patients by evaluating the bone marrow morphology and minimal residual disease. While PET-CT (Positron Emission Tomography-Computed Tomography) was used to evaluate disease status of lymphoma patients. Gastrointestinal symptoms and liver as well as kidney function were evaluated according to CTCAE5.0 criteria.

Statistical analyses

All clinical data were analyzed using SPSS (SPSS, Chicago, IL, USA) and Prism5 (GraphPad Software, La Jolla, CA, USA). Median values and ranges were used to present continuous variables and percentages for categorical variables. Groups were compared using Pearson's Chi-square tests or Fisher's exact test for categorical variables and non-parametric tests for continuous variables when the data did not follow a normal distribution or homogeneity of variance. The Kaplan–Meier method was performed to estimate the rate of OS and EFS. Cox proportional hazards regression models were used to explore the protective factors of OS and EFS, and the results are expressed as hazard ratio (HR) with 95% confidence interval. All statistical tests were two-sided, and statistical significance was set at a p value of <0.05.

Results

Patient characteristics

The median time of treatment was 14.0 (range = 2.0–30.0) and 25.0 (range = 8.0–50.0) days for Ava and BATs groups, respectively.

Table 1 shows the demographic and clinical characteristics of both the groups, and they did not differ in terms of sex, age, and disease type. No significant differences in pretreatment platelets, leukocyte counts, or hemoglobin levels between the groups were observed.

Treatment efficacy

Platelet response. The cumulative incidence of PR within 3 months after enrollment was 75.6% and 13.3% for the Ava and BATs groups, respectively. Most patients in the Ava group achieved PR within 1 month (30/31 patients), and only one patient achieved PR after 1 month (40 days). Two of the four patients in the BATs group achieved PR within 1 month [($p < 0.001$, Figure 2(a)]. The median time to PR was 14.0 (5.0–40.0) and 33.5 (24.0–55.0) days for the Ava and BATs groups, respectively. Among CIT patients, 76.3% (29/38) achieved PR in the Ava group, and 14.8% (4/27) achieved PR in the BATs group [$p < 0.001$, Figure 2(b)]. There were three non-CIT patients in both the groups, of which two patients in the Ava group reached PR, while none of the patients in the BATs group reached PR [Figure 2(b)]. The median platelet counts at baseline (Day 0) did not differ between the two groups [Ava 7.0 (range 1.0–24.0) $\times 10^9$/L, BATs 10.0 (range 2.0–23.0) $\times 10^9$/L; $p = 0.102$]. On days 7, 14, 21, and 28 after enrollment, significantly higher median platelet counts were observed in the Ava group than those in the BATs group [21.0 (interquartile range (IQR) 11.0–35.0) *versus* 10.0 (IQR 5.5–17.5) $\times 10^9$/L, $p = 0.002$; 31.0 (IQR 14.0–92.0) *versus* 10.5 (IQR 5.0–21.0) $\times 10^9$/L, $p = 0.002$; 86.0 (IQR 28.0–234.0) *versus* 18.0 (IQR 4.5–30.0) $\times 10^9$/L, $p < 0.001$, and 144.5 (IQR 59.0–241.0) *versus* 20.0 (IQR 10.0–29.0) $\times 10^9$/L, $p < 0.001$, respectively.] [Figure 2(c)].

Platelet transfusion independence rate and cumulative platelet transfusion units. The rate of platelet transfusion independence on day 30 was 78.0% *versus* 30.0% [Ava group *versus* BATs group, $p < 0.001$, Figure 3(a)]. The median cumulative platelet transfusion units on day 30 of the Ava and BATs groups were 10.0 (range = 1.0–29.0) U and 13.0 (range = 2.0–38.0) U, respectively [$p = 0.033$, Figure 3(b)]. The cumulative incidence of platelet transfusion independence at the end of the follow-up (3 months) was 78.0%

Table 1. Demographic and baseline characteristics of patients.

Characteristic	Patients	Ava group BATs group		*p* Value
		n = 41	*n* = 30	
Median patients age (range)	46 (21–73)	44 (21–68)	52 (25–73)	0.050
Gender, *n* (%)				0.784
Male	22 (31.0)	11 (26.8)	11(36.7)	
Female	49 (69.0)	30 (73.2)	19 (63.3)	
Disease, *n* (%)				0.289
AML	31 (43.7)	18(43.9)	13 (43.3)	
ALL	10 (14.1)	8 (19.5)	2 (6.7)	
MDS	14 (19.7)	5 (12.2)	9 (30.0)	
Lymphoma	5 (7.0)	3 (7.3)	2 (6.7)	
Other	11 (15.5)	7 (17.1)	4 (13.3)	
ABO blood type, *n* (%)				0.492
A	18 (25.4)	13 (31.7)	5 (16.6)	
B	15 (21.1)	7 (17.1)	8 (26.7)	
O	31 (43.7)	17 (41.5)	14 (46.7)	
AB	7 (9.9)	4 (9.7)	3 (10.0)	
Median baseline blood routine (range)				
WBC count (G/L)	2.11 (IQR 0.8–5.7)	1.91 (IQR 0.6–12.3)	2.16 (IQR 0.9–4.0)	0.866
HGB count (G/L)	64 (IQR 55–75)	60 (IQR 55–72.5)	64 (IQR 55–76.2)	0.499
Platelet count (G/L)	8 (IQR 6–12)	7 (IQR 5–11)	10 (IQR 6.75–14)	0.102
CIT, *n* (%)				0.688
Yes	65 (91.5)	38 (92.7)	27 (90.0)	
No	6 (8.5)	3 (7.3)	3 (10.0)	
			BAT treatment, *n* (%)	
			IVIG 27 (75.0)	
			Steroids 5 (13.9)	
			HLA-matched platelet 4 (11.1)	

ALL, acute lymphoblastic leukemia; AML, acute myelocytic leukemia; Ava, Avatrombopag; BAT, best available therapy; CIT, chemotherapy-induced thrombocytopenia; HGB, hemoglobin; HLA, human leucocyte antigen; IQR, interquartile range; IVIG, intravenous immunoglobulins; MDS, myelodysplastic syndrome; Other, including aplastic anemia, primary myelofibrosis, mixed acute leukemia, acute monocytic leukemia, chronic myeloid leukemia, chronic lymphocytic leukemia, NK cellular leukemia, multiple myeloma; PLT, platelet; WBC, white blood cell.

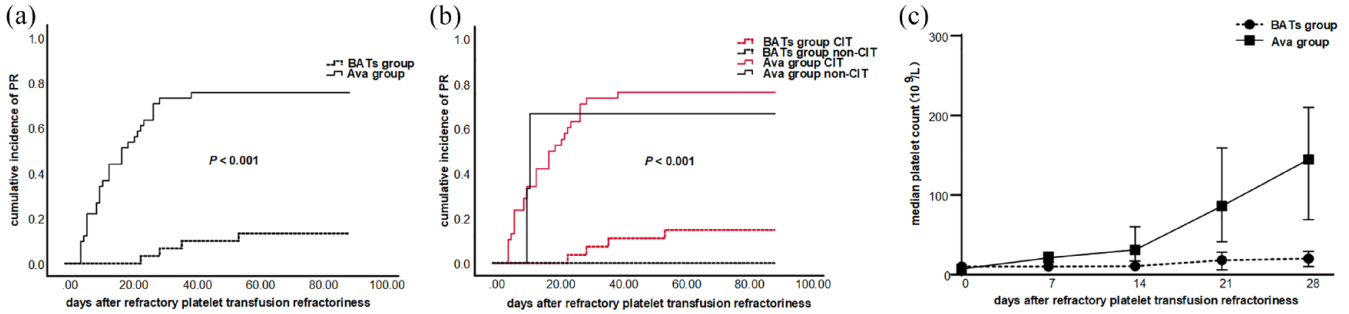

Figure 2. Efficacy of PR between two groups. (a) The cumulative incidence of PR on day 90. (b) The cumulative incidence of PR in both groups with CIT and non-CIT patients, respectively. (c) Differences in median platelet counts between two groups at day 0 ($p = 0.102$), day 7 ($p = 0.002$), day 14 ($p = 0.002$), day 21 ($p < 0.001$), and day 28 ($p < 0.001$), respectively. Continuous line stands for Ava group; dashed line stands for BATs group; error bars represent interquartile ranges.
Ava, Avatrombopag; BAT, best available therapy; CIT, chemotherapy-induced thrombocytopenia; PR, platelet response.

versus 33.3% [Ava group *versus* BATs group, $p < 0.001$, Figure 3(c)]. All the patients reached off-transfusion in the Ava group within 1 month, while one of the nine patients in the BATs group achieved off-transfusion after 1 month (43 days). The median time to platelet transfusion independence was 7.0 (range = 2.0–28.0) *versus* 15.0 (range = 7.0–43.0) days [Ava group *versus* BATs group, $p < 0.001$, Figure 3(d)].

Bleeding outcomes. At baseline, there were 24.4% (10/41) patients with grade 3–4 bleeding in the Ava group and 26.7% (8/30) in the BATs group (24.4% *versus* 26.7%, $p = 0.828$). Further, there were 19.5% (8/41) and 26.7% (8/30) patients with grade 1–2 bleeding in both groups (Ava group *versus* BATs group, respectively, $p = 0.476$). The overall bleeding rate was 43.9% and 53.3%, respectively. No significant statistical differences in the overall bleeding rate between the two groups on baseline were reported [Ava group *versus* BATs group, $p = 0.432$, Figure 4(a) and (b)]. However, WHO grade 3–4 bleeding occurred in 12.2% (5/41) and 56.7% (17/30) cases (56.7%) in the Ava and BATs groups at the end of follow-up, respectively ($p < 0.001$), whereas 9.8% (4/41) and 16.7% (5/30) patients had grade 1–2 bleeding in the Ava and BATs groups, respectively. No statistical differences in grade 1–2 bleeding between the two groups were noted (9.8% *versus* 16.7%, $p = 0.387$). Nevertheless, the rate of overall bleeding in any grade was significantly higher in the BATs group than that in the Ava group [73.3% and 21.9%; $p < 0.001$, Figure 4(a) and (b)].

OS and EFS. In the Ava group, 34 patients survived and 7 died. WHO grade 3 or 4 bleeding occurred in five patients. In the BATs group, 18 patients survived and 12 died. Seventeen patients experienced three to four bleeding events. The 3-month OS was 82.9% *versus* 60.0% [$p = 0.029$, Ava group *versus* BATs group, Figure 5(a)]. The 3-month EFS was 82.9% *versus* 43.3% [$p < 0.001$, Ava group *versus* BATs group, Figure 5(b)]. Patients in the Ava group had better 3-month OS and EFS than those in BATs group. Causes of death included infection, internal hemorrhage, progression, or recurrence of primary disease. Bleeding symptoms included gastrointestinal bleeding, intracranial bleeding, and urinary bleeding. Cox proportional hazards regression analysis revealed that Ava therapy was a protective factor for the OS ($p = 0.051$) and EFS ($p = 0.011$) of PTR patients (Table 2).

Safety and tolerability. Regarding adverse events, rashes occurred in two (4.9%) patients treated with avatrombopag, no patients experienced severe adverse events or relapse of primary disease. Two patients in BATs group developed gastrointestinal symptoms. The gastrointestinal symptoms in AE patients are mild nausea, which was classified as grade 1 according to CTCAE5.0. Furthermore, no patients stopped using avatrombopag due to side effects or intolerability. Among patients treated with Ava, no instances of disease relapse or progression were observed. However, three patients relapsed in the BATs group. Thus, avatrombopag was well tolerated in our study.

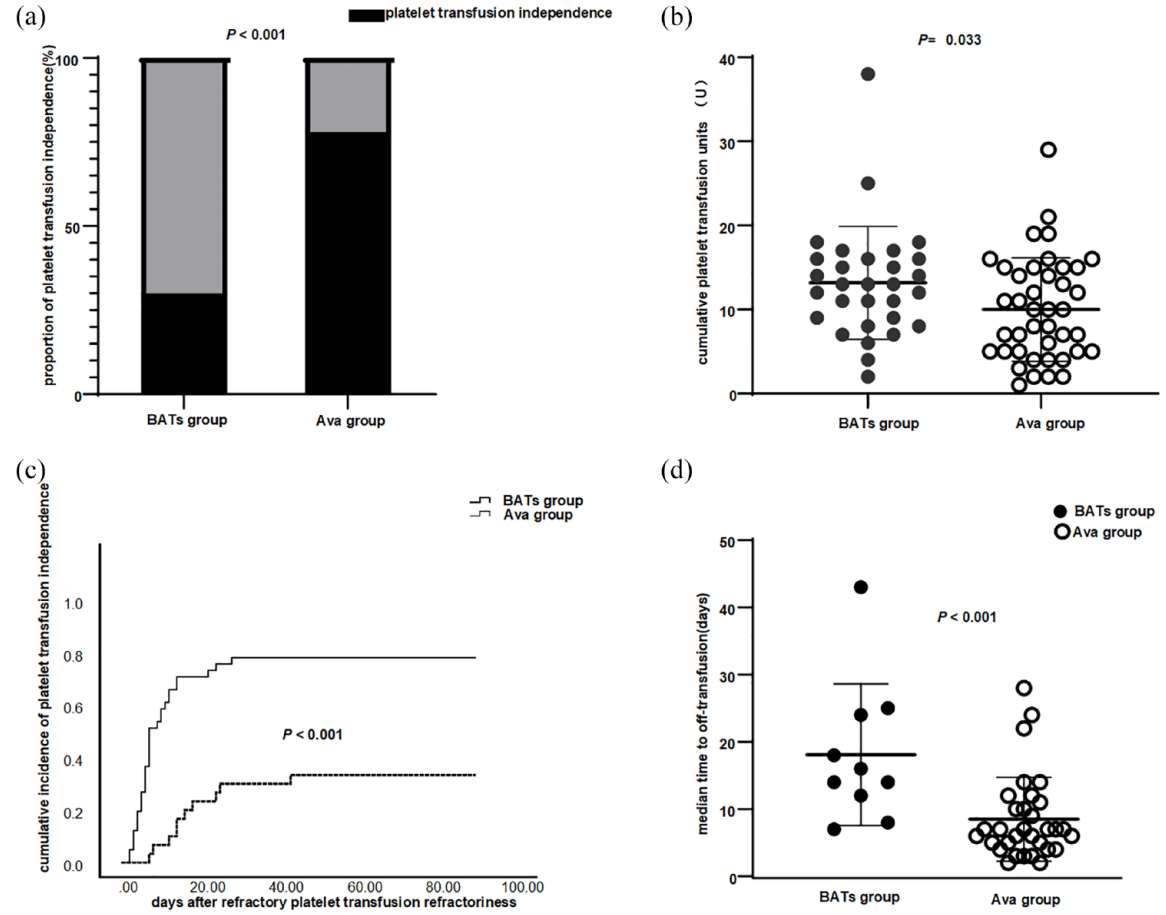

Figure 3. Efficacy on platelet transfusion independence between two groups. (a) The rate of platelet transfusion independence between two groups, Ava group was significantly higher than BATs group on day 30 ($p < 0.001$). (b) The median cumulative platelet transfusion units between two groups, the cumulative platelet transfusion units of Ava group were less than that of BATs group ($p = 0.033$). (c) The cumulative rate of platelet transfusion independence between two groups. At the end of follow-up, the cumulative platelet transfusion independence rate of Ava group was significantly higher than that of BATs group ($p < 0.001$). (d) The median time to platelet transfusion independence between two groups, the median time of disengagement in Ava group was significantly shorter than that in BATs group ($p < 0.001$).
Ava, Avatrombopag; BAT, best available therapy.

Discussion

To the best of our knowledge, this study is the first to report the clinical application of avatrombopag in refractory PTR treatment. Our results showed that this drug exhibited good efficacy and safety profile and improved the OS in patients with refractory PTR. Furthermore, no progression of primary diseases was observed.

Immune factors play an important role in patients with refractory PTR. In fact, the HLA system is identified as the dominating cause of immune-mediated platelet refractoriness.[33] The mechanisms underlying immune-related PTR include antibody-mediated platelet clearance and inhibition of megakaryocyte production and maturation. Experimental data from a mouse model showed that HLA inhibition protects megakaryocytes against HLA antibody-mediated complement-dependent and cell-mediated cytotoxicity, indicating that HLA-mediated cytotoxicity may lead to the destruction of megakaryocytes and platelets.[34] The main mechanism of PTR treatment using steroids and IVIG is their binding to FcR on the platelet surface to reduce macrophage–phagocytosis thrombocytopenia.[19,20] The refractory PTR patients enrolled in our research did not respond to steroid or IVIG treatment. Therefore,

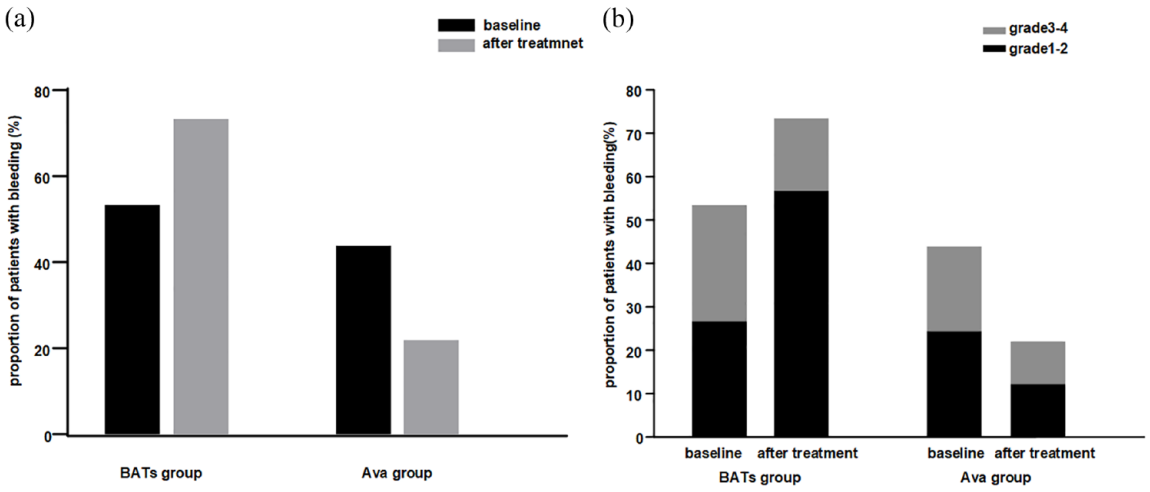

Figure 4. Bleeding outcomes of WHO grades between two groups. (a) On baseline, there were no statistical differences between two groups in terms of the rate of overall bleeding (Ava group *versus* BATs group, $p = 0.432$). At the end of follow-up, rates of overall bleeding in any grades were significantly higher in the BATs group than in the Ava group (73.3% and 21.9%; $p < 0.001$). (b) On baseline, the bleeding rates of grades 1–2 and 3–4 between two groups (Ava group *versus* BATs group, $p = 0.476$, $p = 0.828$, respectively) were not statistical difference. At the end of follow-up, there were no differences in bleeding grade 1–2 between two groups ($p = 0.387$), while patients with bleeding grade 3–4 were significantly higher in the BATs group than those of Ava group ($p < 0.001$).
Ava, Avatrombopag; BAT, best available therapy; WHO, World Health Organization.

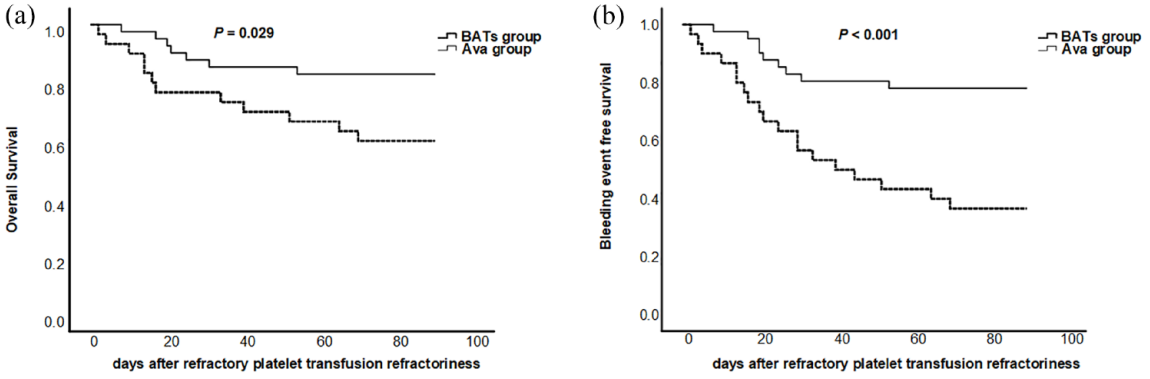

Figure 5. OS and EFS between two groups. (a) OS in patients with refractory PTR. Avatrombopag improved the OS of patients with refractory PTR within 3 months (Ava group *versus* BATs group, $p = 0.029$). (b) Bleeding events free survival in patients with refractory PTR. Avatrombopag improved the EFS of patients with refractory PTR within 3 months (Ava group *versus* BATs group, $p = 0.001$).
Ava, Avatrombopag; BAT, best available therapy; EFS, event-free survival; OS, overall survival; PTR, platelet transfusion refractoriness.

the activation of Fc-mediated phagocytosis of macrophages by anti-HLA antibodies may not be the critical mechanism in these patients. Other mechanisms of immune-related PTR include $CD8^+$ T-cell mediated megakaryocyte apoptosis, maturation defects, and decreased pro-platelet formation.[18] The cytotoxic effects of $CD8^+$ T cells directly impair platelet production when co-cultured with megakaryocytes *in vitro*.[18] Further, HLA antibodies have been reported to inactivate regulatory T cells, leading to a decrease in the levels of TGF-β1.[35,36] However, few clinical therapeutic strategies are currently targeted at T-cell immune modulation. Immunosuppressive drugs such as sirolimus and cyclosporine can inhibit T-cell activation and proliferation, thus

Table 2. Cox proportional hazards regression analysis for OS, and bleeding events free survival.

Variable	OS		Bleeding events free survival	
	HR (95% CI)	*p* Value	HR (95% CI)	*p* Value
Treatment (Ava *versus* BATs)	0.306 (0.108–0.868)	0.026	0.239 (0.091–0.627)	0.004
Age	1.006 (0.966–1.047)	0.784	0.996 (0.961–1.032)	0.825
Gender (female *versus* male)	0.349 (0.11–1.103)	0.073	2.318 (0.792–5.77)	0.134
Disease (AML *versus* MDS)	1.890 (0.546–6.54)	0.315	2.375 (0.761–7.414)	0.137

AML, acute myelocytic leukemia; Ava, Avatrombopag; BAT, best available therapy; CI, confidence interval; HR, hazard ratio; MDS, myelodysplastic syndrome; OS, overall survival.

increasing the number of functional T-reg cells.[37] However, these drugs are slow in exerting their effect and may aggravate bone marrow suppression.[38] Therefore, they are not applicable to PTR patients with malignant diseases. In addition, the strict HLA-matched platelets are difficult to obtain, which limits their application as PTR treatment.[10,11] Recent studies have suggested that TPO-RA may be able to induce immune tolerance in immune thrombocytopenia patients.[39] TPO-RA binds to the thrombopoietin receptors, leading to the activation of the JAK2/STAT5 pathway and facilitation of megakaryocyte proliferation and platelet production.[21,40] A rapid increase in platelet mass induced by TPO-RA increases antigen mass, which leads to subsequent antigen contact and, thereby, suppression of T-cell activation[25,26,41,42] as well as increased circulation levels of TGF-β1.[25,26,28,41–43] Therefore, TPO-RA may exert its beneficial effect in treating refractory PTR through the regulation of $CD8^+$ T cells and circulating TGF-β1 levels. Since the HLA antibody or HPA (human platelet antigen) antibody testing is unobtainable in our hospital, further laboratory investigations are needed to verify this hypothesis.

TPO-RA induces the phosphorylation of platelet proteins and mediates platelet aggregation and activation through c-MPL receptors. In addition, TPO-RA stimulation of platelets causes functional modulation of integrins, induction of fibrinogen binding, and subsequent platelet aggregation.[44,45] Krečak *et al.*[46] reported a successful case of using TPO-RA treatment for immune-mediated refractoriness in severe thrombocytopenia associated with MDS. The platelet count was improved and maintained at $32–65 \times 10^9$/L upon admission of eltrombopag and subsequent with romiplostim. Study also indicated the favorable efficacy of TPO-RA in enhancing platelet activation and improving hemostasis, irrespective of platelet count. In our study, 12 patients in Ava group did not reach PR; however, four patients experienced relief in their bleeding symptoms. While 26 patients did not achieve PR in the BATs group, only 2 cases attained relief in bleeding symptoms. Our findings indicated that avatrombopag treatment not only increased the platelet count but also reduced the severity of bleeding symptoms in refractory PTR patients. A possible explanation may be that Ava could promote platelet aggregation and enhance platelet attachment to collagen, regardless of the platelet count.

A previous study showed that PTR was significantly associated with early mortality. The 100-day survival rates were 98% and 83% ($p < 0.01$) in non-PTR and PTR patients, respectively.[47] This is consistent with our data, which suggested that the response outcomes of Ava also translate into survival advantages in patients with refractory PTR.

The application of TPO-RA in malignant hematological diseases remains controversial, with anxiety on the clonal evolution of AML, MDS, and SAA (serious-aplastic-anemia) patients.[48–50] The main concern is that cytokines stimulate self-renewal and proliferation of hematopoietic stem and progenitor cells, which may increase the risk of clonal evolution and consequent malignant transformation.[48] However, other studies have suggested an opposite effect.[22,51,52] Will *et al.* investigated the effects of TPO-RA on the proliferation, apoptosis, differentiation, colony formation, and malignant self-renewal of mononuclear cells from the bone marrow of AML and MDS

patients. They reported that malignant mononuclear cells did not exhibit increased proliferative or clonogenic capacity,[22] while TPO-RA might promote normal megakaryocytic colony formation and megakaryocytic differentiation in AML and MDS patients.[22] Our study showed that no instances of disease relapse or progression were observed in Ava group within 3 months. However, the follow-up period is too short to make the conclusion. The potential impact of avatrombopag on malignant disease status still requires long-term follow-up observation.

Limitation

The conclusion of this study was not fully impartial since it was retrospective, relatively small sample size, and short observation period. Therefore, more extensive studies are required to deeply understand the role and efficacy of this drug for refractory PTR, further explore the mechanisms underlying this disease and optimize the therapeutic approach.

Conclusion

In summary, although the treatment of refractory PTR is challenging, our study demonstrated that it may be improved and become less refractory since the emergence of avatrombopag. Moreover, thanks to the easy availability and good tolerability of avatrombopag, it can become a complementary treatment option for HLA-typed platelets.

Declarations

Ethics approval and consent to participate

The study was approved by the Institutional Ethics Committee of Nanfang Hospital, Southern Medical University (No. NFEC-2023-065). The requirement to obtain informed consent was waived due to the retrospective nature of the study and was deemed exempt from review by the Ethics Committee of the Nanfang Hospital of Southern Medical University.

Consent for publication

Not applicable.

Author contributions

Yuehong Qin: Conceptualization; Formal analysis; Writing – original draft; Writing – review & editing.

Yu Wang: Data curation; Formal analysis; Investigation; Methodology.

Yujiao Zhang: Formal analysis; Methodology; Software.

Yingying Jiao: Data curation; Formal analysis.

Jieyu Ye: Conceptualization; Funding acquisition; Methodology; Resources; Supervision; Validation; Writing – review & editing.

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Competing interests

All authors declare no competing interests.

Availability of data and materials

Not applicable.

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References

1. Cohn CS. Platelet transfusion refractoriness: how do I diagnose and manage? *Hematol Am Soc Hematol Educ Program* 2020; 2020: 527–532.
2. Hod E and Schwartz J. Platelet transfusion refractoriness. *Br J Haematol* 2008; 142: 348–360.
3. Trial to Reduce Alloimmunization to Platelets Study Group. Leukocyte reduction and ultraviolet B irradiation of platelets to prevent alloimmunization and refractoriness to platelet transfusions. *N Engl J Med* 1997; 337: 1861–1869.
4. Hu X, Cai H, Zheng L, *et al.* Clinical and immunological features of platelet transfusion refractoriness in young patients with *de novo*

acute myeloid leukemia. *Cancer Med* 2020; 9: 4941–4948.

5. Slichter SJ, Davis K, Enright H, *et al.* Factors affecting posttransfusion platelet increments, platelet refractoriness, and platelet transfusion intervals in thrombocytopenic patients. *Blood* 2005; 105: 4106–4114.
6. Tanoue S, Konuma T, Kato S, *et al.* Platelet transfusion refractoriness in single-unit cord blood transplantation for adults: risk factors and clinical outcomes. *Biol Blood Marrow Transplant* 2018; 24: 1873–1880.
7. Gurevich-Shapiro A, Tzadok S, Rosenberg A, *et al.* Extended small-dose platelet transfusions in multitransfused hemato-oncological patients: a single-center experience. *Acta Haematol* 2017; 137: 183–190.
8. Terrault N, Chen Y-C, Izumi N, *et al.* Avatrombopag before procedures reduces need for platelet transfusion in patients with chronic liver disease and thrombocytopenia. *Gastroenterology* 2018; 155: 705–718.
9. Bussel JB, Kuter DJ, Aledort LM, *et al.* A randomized trial of avatrombopag, an investigational thrombopoietin-receptor agonist, in persistent and chronic immune thrombocytopenia. *Blood* 2014; 123: 3887–3894.
10. Stanworth SJ, Navarrete C, Estcourt L, *et al.* Platelet refractoriness – practical approaches and ongoing dilemmas in patient management. *Br J Haematol* 2015; 171: 297–305.
11. Schmidt AE, Refaai MA and Coppage M. HLA-mediated platelet refractoriness. *Am J Clin Pathol* 2019; 151: 353–363.
12. Liu W, Wu D, Hu T, *et al.* Efficiency of treatment with rituximab in platelet transfusion refractoriness: a study of 7 cases. *Int J Clin Exp Med* 2015; 8: 14080–14084.
13. Mauro M, Camoglio F, Piccoli P, *et al.* The use of splenectomy to manage platelet transfusion refractoriness due to anti-human leukocyte antibodies in allogeneic stem cell transplantation. *Pediatr Rep* 2016; 8: 6159.
14. Cid J, Magnano L, Acosta M, *et al.* Rituximab, plasma exchange and intravenous immunoglobulins as a new treatment strategy for severe HLA alloimmune platelet refractoriness. *Platelets* 2015; 26: 190–194.
15. Miki H, Ozaki S, Tanaka O, *et al.* Marked improvement of platelet transfusion refractoriness after bortezomib therapy in multiple myeloma. *Int J Hematol* 2009; 89: 223–226.
16. Meinke S, Karlström C and Höglund P. Complement as an immune barrier in platelet transfusion refractoriness. *Transfus Med Rev* 2019; 33: 231–235.
17. Aslam R, Kapur R, Segel GB, *et al.* The spleen dictates platelet destruction, anti-platelet antibody production, and lymphocyte distribution patterns in a murine model of immune thrombocytopenia. *Exp Hematol* 2016; 44: 924–930.e1.
18. Chow L, Aslam R, Speck ER, *et al.* A murine model of severe immune thrombocytopenia is induced by antibody- and CD8+ T cell-mediated responses that are differentially sensitive to therapy. *Blood* 2010; 115: 1247–1253.
19. Nagelkerke SQ, Porcelijn L, Geissler J, *et al.* The association and functional relevance of genetic variation in low-to-medium-affinity Fc-gamma receptors with clinical platelet transfusion refractoriness. *J Thromb Haemost* 2020; 18: 2047–2053.
20. Witkowski M, Witkowska M and Robak T. Autoimmune thrombocytopenia: current treatment options in adults with a focus on novel drugs. *Eur J Haematol* 2019; 103: 531–541.
21. Erickson-Miller CL, Delorme E, Tian SS, *et al.* Preclinical activity of eltrombopag (SB-497115), an oral, nonpeptide thrombopoietin receptor agonist. *Stem Cells (Dayton, Ohio)* 2009; 27: 424–430.
22. Will B, Kawahara M, Luciano JP, *et al.* Effect of the nonpeptide thrombopoietin receptor agonist Eltrombopag on bone marrow cells from patients with acute myeloid leukemia and myelodysplastic syndrome. *Blood* 2009; 114: 3899–3908.
23. Kuter DJ. The structure, function, and clinical use of the thrombopoietin receptor agonist avatrombopag. *Blood Rev* 2022; 53: 100909.
24. Gilreath J, Lo M and Bubalo J. Thrombopoietin receptor agonists (TPO-RAs): drug class considerations for pharmacists. *Drugs* 2021; 81: 1285–1305.
25. Viola A and Lanzavecchia A. T cell activation determined by T cell receptor number and tunable thresholds. *Science (New York, NY)* 1996; 273: 104–106.
26. Schifferli A and Kühne T. Thrombopoietin receptor agonists: a new immune modulatory strategy in immune thrombocytopenia? *Semin Hematol* 2016; 53(Suppl. 1): S31–S34.
27. Sadallah S, Eken C and Schifferli JA. Ectosomes as immunomodulators. *Semin Immunopathol* 2011; 33: 487–495.

28. Zheng SG. The critical role of TGF-beta1 in the development of induced Foxp3+ regulatory T cells. *Int J Clin Exp Med* 2008; 1: 192–202.

29. Nishimoto T, Numajiri M, Nakazaki H, *et al.* Induction of immune tolerance to platelet antigen by short-term thrombopoietin treatment in a mouse model of immune thrombocytopenia. *Int J Hematol* 2014; 100: 341–344.

30. Cui Y, He Y, Hu C, *et al.* Avatrombopag for the treatment of thrombocytopenia induced by chemotherapy in patients with solid tumors: a multicenter, open-label, single-arm trial. *Front Pharmacol* 2022; 13: 970978.

31. Drozd-Sokolowska JE and Wiktor-Jedrzejczak W. Factors determining the risk of severe (WHO grades 3 and 4) hemorrhage in hematologic patients. *Transfus Apher Sci* 2011; 44: 129–134.

32. National Cancer Institute. Common terminology criteria for adverse events (CTCAE). Version 5.0. https://ctepcancergov/protocoldevelopment/electronic_applications/ctchtm#ctc_50 (2017).

33. Prodger CF, Rampotas A, Estcourt LJ, *et al.* Platelet transfusion: alloimmunization and refractoriness. *Semin Hematol* 2020; 57: 92–99.

34. Gras C, Schulze K, Goudeva L, *et al.* HLA-universal platelet transfusions prevent platelet refractoriness in a mouse model. *Hum Gene Ther* 2013; 24: 1018–1028.

35. Turner JA, Stephen-Victor E, Wang S, *et al.* Regulatory T cell-derived TGF-β1 controls multiple checkpoints governing allergy and autoimmunity. *Immunity* 2020; 53: 1202–1214.e6.

36. Melief CJM and Kessler JH. Novel insights into the HLA class I immunopeptidome and T-cell immunosurveillance. *Genome Med* 2017; 9: 44.

37. Wu C, Wang Q, Xu D, *et al.* Sirolimus for patients with connective tissue disease-related refractory thrombocytopenia: a single-arm, open-label clinical trial. *Rheumatology (Oxford, England)* 2021; 60: 2629–2634.

38. Saris A and Pavenski K. Human leukocyte antigen alloimmunization and alloimmune platelet refractoriness. *Transfus Med Rev* 2020; 34: 250–257.

39. Bao W, Bussel JB, Heck S, *et al.* Improved regulatory T-cell activity in patients with chronic immune thrombocytopenia treated with thrombopoietic agents. *Blood* 2010; 116: 4639–4645.

40. Broudy VC and Lin NL. AMG531 stimulates megakaryopoiesis *in vitro* by binding to MPL. *Cytokine* 2004; 25: 52–60.

41. Taams LS, van Eden W and Wauben MH. Dose-dependent induction of distinct anergic phenotypes: multiple levels of T cell anergy. *J Immunol (Baltimore, MD)* 1999; 162: 1974–1981.

42. Ferber I, Schönrich G, Schenkel J, *et al.* Levels of peripheral T cell tolerance induced by different doses of tolerogen. *Science (New York, NY)* 1994; 263: 674–676.

43. Korn T, Bettelli E, Oukka M, *et al.* IL-17 and Th17 cells. *Annu Rev Immunol* 2009; 27: 485–517.

44. Chen J, Herceg-Harjacek L, Groopman JE, *et al.* Regulation of platelet activation *in vitro* by the c-MPL ligand, thrombopoietin. *Blood* 1995; 86: 4054–4062.

45. van Dijk WEM, Brandwijk ON, Heitink-Polle KMJ, *et al.* Hemostatic changes by thrombopoietin-receptor agonists in immune thrombocytopenia patients. *Blood Rev* 2021; 47: 100774.

46. Krečak I, Skorić I, Čengić M, *et al.* Immune-mediated platelet transfusion refractoriness in a severely thrombocytopenic patient with myelodysplastic syndrome successfully treated with romiplostim. *Transfus Apher Sci* 2023; 62: 103582.

47. Kerkhoffs JL, Eikenboom JC, van de Watering LM, *et al.* The clinical impact of platelet refractoriness: correlation with bleeding and survival. *Transfusion* 2008; 48: 1959–1965.

48. Winkler T, Fan X, Cooper J, *et al.* Treatment optimization and genomic outcomes in refractory severe aplastic anemia treated with eltrombopag. *Blood* 2019; 133: 2575–2585.

49. Luo SS, Ogata K, Yokose N, *et al.* Effect of thrombopoietin on proliferation of blasts from patients with myelodysplastic syndromes. *Stem Cells (Dayton, Ohio)* 2000; 18: 112–119.

50. Corazza F, Hermans C, D'Hondt S, *et al.* Circulating thrombopoietin as an *in vivo* growth factor for blast cells in acute myeloid leukemia. *Blood* 2006; 107: 2525–2530.

51. Zhou M, Qi J, Gu C, *et al.* Avatrombopag for the treatment of thrombocytopenia post hematopoietic stem-cell transplantation. *Ther Adv Hematol* 2022; 13: 20406207221127532.

52. Roth M, Will B, Simkin G, *et al.* Eltrombopag inhibits the proliferation of leukemia cells via reduction of intracellular iron and induction of differentiation. *Blood* 2012; 120: 386–394.