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ORIGINAL RESEARCH

Porcine Anti-Lymphocyte Globulin, Cyclosporine A Plus Thrombopoietin Receptor Agonists Achieved Similar Efficacy and Survival Compared to Allogeneic Hematopoietic Stem Cell Transplantation in Patients with Aplastic Anemia

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Background: Immunosuppressive therapy (IST) with horse or rabbit anti-human thymocyte immunoglobulin (h-/r-ATG) and hematopoietic stem cell transplantation (HSCT) are two baseline treatments for severe aplastic anemia (SAA) and transfusion-dependent non-severe aplastic anemia (TD-NSAA) patients. Addition of thrombopoietin receptor agonists (TPO-RAs) to standard IST therapy (h-/r-ATG) has greatly improved the survival of SAA, whereas porcine anti-lymphocyte globulin (p-ALG) combined with TPO-RAs still had a matter of debate.

Methods: We retrospectively compared the data of 48 AA patients in our center between 2020 and 2022, 23 AA patients received with p-ALG ± TPO-RAs, 25 AA patients underwent matched sibling donor (MSD-) or haploidentical (haplo-) HSCT.

Results: For patients in the HSCT group, the ORR was 90.9% which was significantly higher than that in the IST±TPO-RAs group (45.5%, $P = 0.001$) at 3 months; moreover, patients who underwent HSCT achieved faster transfusion independence, better CR rate, shorter time of recovery normal blood routine, and the percentage of normal blood routine (all $P < 0.05$) compared with IST±TPO-RAs group. However, the ORR were similary at 6 months in the two groups (95.5% vs 81.8% $P = 0.342$), with a median follow up of 19.8 months (range, 0.3–38.2 months), the 2-year FFS and OS in the two cohorts has no different. Subgroup analysis further indicated that the 2-year FFS and OS were similar between IST+TPO-RAs and haplo-HSCT subgroups, as well as in IST+TPO-RAs and MSD-HSCT cohorts. Moreover, the first-time hospitalizations were much more expensive in the HSCT group than in the IST±TPO-RAs group (402 756 vs. 292 902 yuan, $P = 0.002$).

Conclusion: P-ALG-based-IST±TPO-RAs is a good treatment option with similar FFS and OS compared to allo- HSCT for AA patients without the opportunity of HSCT.

Keywords: porcine anti-human thymocyte immunoglobulin, thrombopoietin receptor agonist, hematopoietic stem cell transplantation, aplastic anemia

Introduction

Acquired aplastic anemia (AA) is a rare syndrome of bone marrow failure caused by immune dysfunction of different etiologies which destroy hematopoietic stem cell clones, with clinical manifestations of anemia, infection, and bleeding.[1] The guidelines of the British Society in 2016 and Chinese in 2022 recommend that first-line treatment of SAA and TD-NSAA is divided by age, with matched sibling hematopoietic stem cell transplantation (MSD-HSCT) preferred for younger patients and immunosuppressive therapy (IST) for elderly patients or those without transplant conditions.[2,3]

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IST is cyclosporine A (CsA) combined with anti-human thymocyte immunoglobulin (ATG) which currently encompasses horse ATG (h-ATG), rabbit ATG (r-ATG) and porcine ATG (p-ALG). H-/r-ATG are widely used in Europe and the United States, and only p-ALG has been approved in China. Studies have suggested that p-ALG had similar or even better outcomes to r-ATG.[4–6] Ma et al[5] retrospectively compared the efficacy and survival of untreated SAA patients who used pALG or rATG. The 1-year ORR was 83.78% for p-ALG and 66.67% for r-ATG ($P = 0.036$); the 5-year OS was 82.22% for p-ALG group and 68.75% for r-ATG group ($P = 0.32$). Chen et al[4] retrospectively compared p-ALG and r-ATG in Chinese patients with treatment-naïve SAA. The 3-month ORR was higher in the p-ALG than that in the r-ATG group (65.8% vs 44.1%, P=0.023), but the ORR reached 74.6% and 64.7% ($P = 0.361$) after six months. Besides, the minimum number of lymphocytes was significantly lower in the r-ATG group than that in the p-ALG group at 1st-, 3rd-, and 6th-month after therapy. The incidence of myelodysplastic syndrome/acute myeloid leukemia (MDS/AML) transformation significantly lower in the p-ALG than in the r-ATG groups (2.6% vs 11.8%, $P = 0.049$).

Thrombopoietin receptor agonists (TPO-RAs), including eltrombopag, herombopag, and avatrombopag, added to standard IST have been shown to improve the speed and depth of a hematological response in patients with SAA.[7] The combination of IST, mainly using r-ATG, and eltrombopag (EPAG) at up-front treatment in patients with SAA is no different in OS but has an inferior FFS than MSD-HSCT.[8] However, no study has been directly compared p-ALG-based IST with HSCT. Thus, we retrospect the data in our center to find the efficacy and economic outcomes between p-ALG, CsA combined with TPO-RAs and HSCT in patients with AA.

Patients and Methods

Patients

Forty-eight patients diagnosed with AA who received MSD-/haplo-HSCT or IST (patients used p-ALG) at the First Affiliated Hospital of Nanchang University between March 2020 and December 2022 were enrolled. Very severe aplastic anemia (VSAA): for patients meeting the criteria for SAA[2] and had an absolute neutrophil count (ANC) below 0.2×10^9/L; patients did not meeting the criteria of SAA but are transfusion dependent was TD-NSAA. Excluding criteria including patients who had congenital bone marrow disorders (Fanconi anemia, Diamond-Blackfan anemia, and dyskeratosis congenital (DKC)), MDS, and LGL. The protocol of this clinical observation study followed the Declaration of Helsinki and was approved by the Ethics Committee of the First Affiliated Hospital of Nanchang University (IIT2023328). Considering the patient's age and wishes, the results of HLA matching, and disease conditions, the choice of the treatment regimens was carried out mainly in accordance with guidelines. If patients in good condition and has a matching sibling donor or a haploidentical donor, MSD-/haplo-HSCT was administered with their consent; otherwise, IST±TPO-RA was chosen. All patients and donors provided written informed consent for this protocol. For patients younger than 18 years old in the cohort, the consent was carried out by their parents.

Immunosuppressive Therapy ± Thrombopoietin Receptor Agonists

The IST group has 23 patients who lack a suitable matched sibling donor or are unsuitable for transplantation due to patient selection, advanced age, or socioeconomic factors, including 18 used IST+TPO-RAs. IST: Porcine antihuman lymphocyte immunoglobulin (p-ALG) 25 $mg \cdot kg^{-1}$ for 5 days, and oral CsA (3–5 mg/kg/day) over the same time period. CsA was administered for 12–24 months with adjustment of the dose to achieve a whole blood trough concentration of 150–250 μg/L. For prevention of allergic reactions, prednisone was given prior to the first p-ALG, and given the next 10 days and rapidly tapered over the next 1–2 weeks. TPO-RAs, including eltrombopag (from 25 mg, up to 75 mg), herombopag (from 2.5 mg, up to 12.5 mg), and avatrombopag (from 20 mg, up to 40 mg), were chosen according to their choice and taken at least for 6 months. All patients received prophylactic antibiotics, antifungal, and anti-viral therapy according to local policies.

Hematopoietic Stem Cell Transplantation

For MSD-HSCT patients, conditioning regimens including: 1) cyclophosphamide (CY) + r-ATG (CY 200$mg \cdot kg^{-1}$, for 4 days; r-ATG 10$mg \cdot kg^{-1}$, for 4 days); 2) fludarabine (Flu) + Cy + p-ALG (Flu 120$mg \cdot m^{-2}$, for 3 days; CY 90$mg \cdot kg^{-1}$, for

3 days; p-ATG 30mg·kg^{-1}, for 4 days).[9] For patients received haplo-HSCT, modified BuCy+r-ATG (Bu 6.4mg·kg^{-1}, for 2 days; CY 200mg·kg^{-1}, for 4 days; r-ATG 10mg·kg^{-1}, for 4 days) was used as conditioning regimens.

Stem cell from donor were mobilized by continuous subcutaneous injection of 5–10 mg/(kg·d) rhG-CSF for 4–5 days. The total target mononuclear cell count and CD34+ cells were ≥2×10^8/kg and ≥2 × 10^6/kg of the recipient's weight, respectively.

CsA, mycophenolate mofetil, and short-course methotrexate were used as prophylaxis against GVHD.[10] Referring to international criteria,[11,12] acute GVHD (aGVHD) and chronic GVHD (cGVHD) were diagnosed and classified. All patients received prophylactic antibiotics, antifungal, and anti-viral therapy according to local policies.

Evaluations and Following Up

Hemoglobin count >100g/L, neutrophil count >1.5 × 10^9/L, and platelet count >100 × 10^9/L and transfusion-independent were defined as complete response (CR). Patients who did not meet the criteria for SAA and having transfusion-independent were defined as partial response (PR). As for TD-NSAA, PR was characterized by no longer requiring transfusions or at least one lineage has a twofold increase or normalization. No response (NR) was defined as still meeting SAA diagnostic criteria (for SAA) or remained transfusion-dependent (for TD-NSAA). Both CR and PR are considered effective. Early death was defined as death that occurs within 60 days of receiving treatment. Death, NR by 6 months, relapse, disease progression, clonal evolution, or evolution were considered treatment failures for IST; death, engraftment failure, delayed rejection, secondary malignancy, and relapse were considered treatment failures for HSCT. Delayed rejection was defined as primary engraftment followed by at least 2 cell lines of cytopenia with obviously hypocellular BM and without moderate to severe aGVHD. The severity of adverse events was based on Common Terminology Criteria for Adverse Events (CTCAE - Version 5.0). The last follow-up date for all surviving patients was June 5, 2023.

Statistical Analysis

Statistical analyses were performed using SPSS 26.0 software (IBM SPSS) or GraphPad Prism 8. Patient characteristics were compared using the chi-square test, Fisher's exact test, and the nonparametric test for continuous variables. The probabilities of OS and FFS were calculated using the Kaplan–Meier method and compared using the Log rank test. In order to match MSD-HSCT and IST+TPO-RAs, propensity score matching was conducted. All P values were two-sided. The level of statistical significance was defined as $P < 0.05$.

Results

Patient Characteristics

Forty-eight patients with AA were enrolled. Figure 1 shows the enrollment for all the patients in this study. Twenty-five and 23 patients in the HSCT and IST±TPO-RAs groups, respectively. In the HSCT group, 16 patients received MSD-HSCT and 9 received haplo-HSCT. Eighteen patients received IST+TPO-RAs, and five patients received IST alone in IST±TPO-RAs group. Sixteen (64%) patients in the HSCT group and 13 (56.5%) patients in the IST±TPO-RAs group received previous treatment which mostly consisted of CsA and/or androgen. The patient characteristics are shown in Table 1 and Supplement Table 1. From diagnosis to therapy, patients in the HSCT group took longer (median 0.97 months) than those in the IST±TPO-RAs group (median 0.70 months) which may be related to the longer preparation of HSCT. No significant difference was found in the remaining base line characteristics.

Efficacy

A total of 22 patients in each group were evaluated for response. The CR rates of HSCT group were higher in the 3rd (54.5% vs 0%, $P < 0.001$), 6th (63.6% vs 36.4%, $P = 0.051$), 9th (73.7% vs 33.3%, $P = 0.016$), and 12th (88.2% vs 35.3%, $P = 0.004$) month after treatment (Figure 2). In addition, the ORR was 90.9% in the HSCT group, and significantly higher than that in the IST±TPO-RAs group (45.5%, $P = 0.001$) at 3 months, However, the ORR were similar at 6, 9, and 12 months (Table 2) between the two groups.

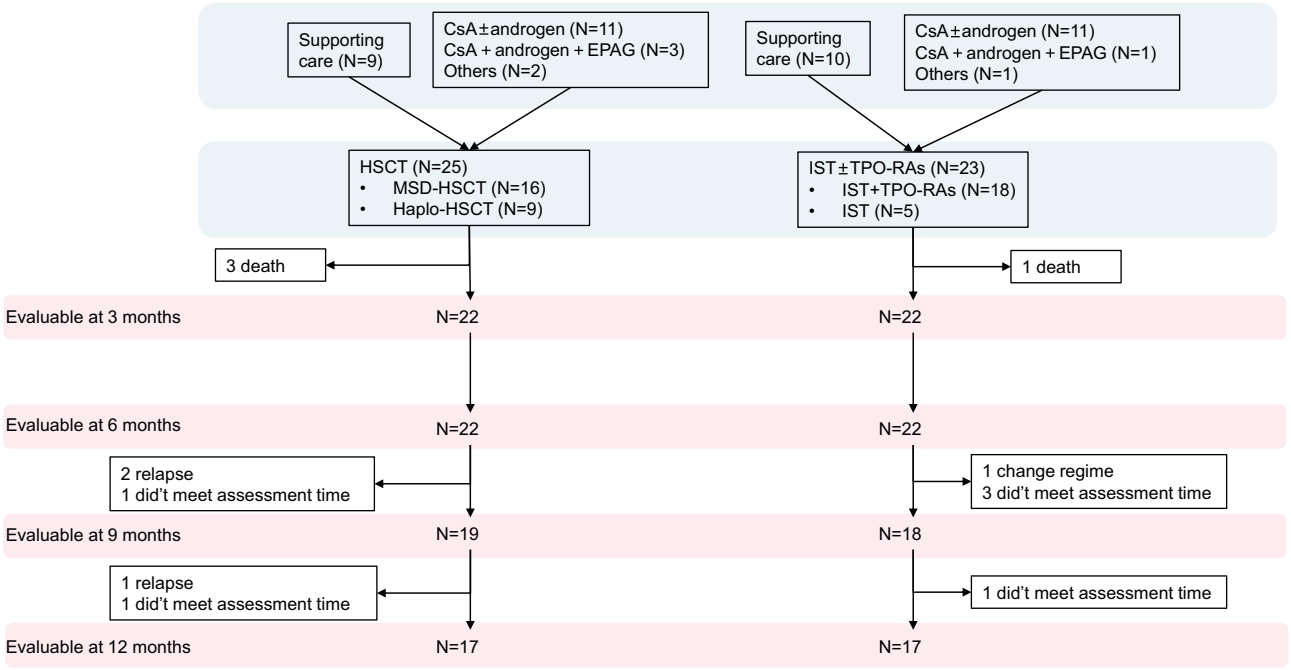

Figure 1 Enrollment flow diagram. A total of 48 patients were enrolled. There were 25 and 23 patients in the HSCT and IST±TPO-RAs groups, respectively. In the HSCT group, 16 patients received MSD-HSCT and 9 received haplo-HSCT. Eighteen patients received IST+TPO-RAs, and five patients received IST alone in IST±TPO-RAs group.

Among survival patients in the IST±TPO-RAs and HSCT group, the median follow-up was 25.0 (range, 5.9–38.2) and 18.8 (range, 5.5–28.2) months, respectively. Twelve (54.5%) and 20 (90.9%) patients finally recovered normal blood routine in the IST±TPO-RAs and HSCT group, respectively. The median time to an ANC $\geq 1.0 \times 10^9$/L were 14 (range 1–85) and 22 (range 17–30) days, respectively, which were similar in the two groups(P=0.924). In the HSCT group the median time to PLT $\geq 100 \times 10^9$/L was shorter than in the IST±TPO-RAs group (14 days vs 33 days, $P < 0.001$) (Table 1). Patients in the HSCT group were weaned from platelet (43 vs 18 days, $P < 0.001$) and erythrocyte (41 vs 19 days, $P < 0.001$) transfusions for a shorter period compared with the IST ± TPO-RAs group.

Table 1 Characteristics of Patients and Clinical Outcomes Between the Two Groups

Variables	HSCT(n=25)	IST±TPO-RAs(n=23)	P value
Median age, year (range)	23(8–60)	28(14–56)	0.285
Age, no. (%)			0.122
< 20 year	12(48)	6(26.1)	
20–40 year	7(28)	8(34.8)	
≥ 40 year	6(24)	9(39.1)	
Male/Female, n (%)	13(52)/12(48)	15(65.2)/8(34.8)	0.359
Disease status, n (%)			0.898
SAA	10(40)	10(43.5)	
vSAA	13(52)	12(52.2)	
TD-NSAA	2(8)	1(4.3)	
Blood routine at diagnosis, median (range)			
Hemoglobin, g/L	79(53–121)	62(29–118)	0.082
PLT, × 10^9/L	8(1–52)	8(1–50)	0.528
ANC, × 10^9/L	0.18(0–1.33)	0.2(0.02–0.78)	0.397
RET, × 10^9/L	8.4(0.9–34.7)	12.4(1.8–68.6)	0. 506

(*Continued*)

Table 1 (Continued).

Variables	HSCT(n=25)	IST±TPO-RAs(n=23)	P value
Median time from diagnosis to treatment, month (range)	0.97(0.23–182.37)	0.70(0.17–32.13)	**0.031**
Previous treatment, n (%)			0.716
Supporting care*	9(36)	10(43.5)	
CsA±androgen	11(44)	11(47.8)	
CsA+androgen +TPO-RA[#]	3(12)	1(4.3)	
Others	2(8)	1(4.3)	
Early death, n (%)	3(12)	1(4.3)	0.663
Donor-recipient sex match, n (%)			–
Female-female	5(20)	–	
Female-male	7(28)	–	
Male-female	7(28)	–	
Male-male	6(24)	–	
HLA-match, n (%)			
5/10	6(24)	–	
6/10	1(4)	–	
7/10	2(8)	–	
10/10	16(64)	–	
Mononuclear cells, $\times 10^8$/kg, median (range)	17.5(11.1–24.3)	–	–
CD34+ cells, $\times 10^6$/kg, median (range)	5.3(2.0–13.6)	–	–
Day +28 myeloid engraftment, n (%)	22(88)	–	–
Median myeloid engraftment, day(range)	14(10–22)	–	–
Day +28 PLT engraftment, n (%)	20(80)	–	–
Median PLT engraftment, day(range)	16(11–110)	–	–
For surviving patients, n (%)			
Median follow-up time, month (range)	18.5(5.5–28.2)	25.0(5.9–38.2)	0.167
Patients with normal blood routine, n (%)	20(90.9)	12(54.5)	**0.007**
Relapsed, n (%)	3(14.3)	2(10)	1.000
Median time to an ANC ≥ 1.0 × 10^9/L, day (range)	22 (17–30)	14(1–85)	0.924
Median time to an PLT≥ 100 × 10^9/L, day (range)	33 (21–242)	179(129–447)	**0.000**
Median time to transfusion independence for platelets, day (range)	18(15–101)	43(14–157)	**0.000**
Median time to transfusion independence for RBCs, day (range)	19(0–88)	41(19–151)	**0.000**
Median first-time hospital stays, day(range)	81(31–156)	75(24–179)	0.569
Median first-time hospitalization expenses, yuan (range)	402,756(200,116–808,954)	292,902(113,865–435,174)	**0.002**

Notes: *Including patients who used G-CSF, TPO, or interleukin-11. [#]Only eltrombopag was used. – Inapplicability. Two-tailed P values <0.05 were considered statistically significant. Bold values indicate statistical significance.

Abbreviations: SAA, severe aplastic anemia; vSAA, very severe aplastic anemia; TD-NSAA, Transfusion-dependent non-severe aplastic anemia; PLT, platelet count; ANC, absolute neutrophil count; RET, reticulocyte; CsA, cyclosporine A; HLA, human leukocyte antigen.

Blood tests revealed significant improvement in both groups at 1, 3, and 6 months following treatment. At one month after therapy, changes in ANC, hemoglobin, platelets, and lymphocytes were all discernible (Figure 3A–D). In the HSCT group, neutrophil, hemoglobin, and platelet counts were 2.47 (0.05–15.17) $\times 10^9$/L, 82 (69–103) g/L, and 83 (5–278) $\times 10^9$/L, which were substantially higher than 1.33 (0.01–2.71) × 10^9/L, 64 (39–89) g/L, and 16 (3–44) × 10^9/L in the IST ±TPO-RAs group. At one month following treatment, the median absolute lymphocyte counts in the HSCT group was 0.24 (0.05–1.33) $\times 10^9$/L, which was lower than the 1.14 (0.04–4.19) $\times 10^9$/L in the IST±TPO-RAs group (Figure 3A). At the third or sixth month, the ANC, hemoglobin, and lymphocytes were essentially the same. It is important to note that the HSCT group's platelets were considerably higher than those in the IST±TPO-RAs group at 1, 3, and 6 months after therapy (Figure 3B).

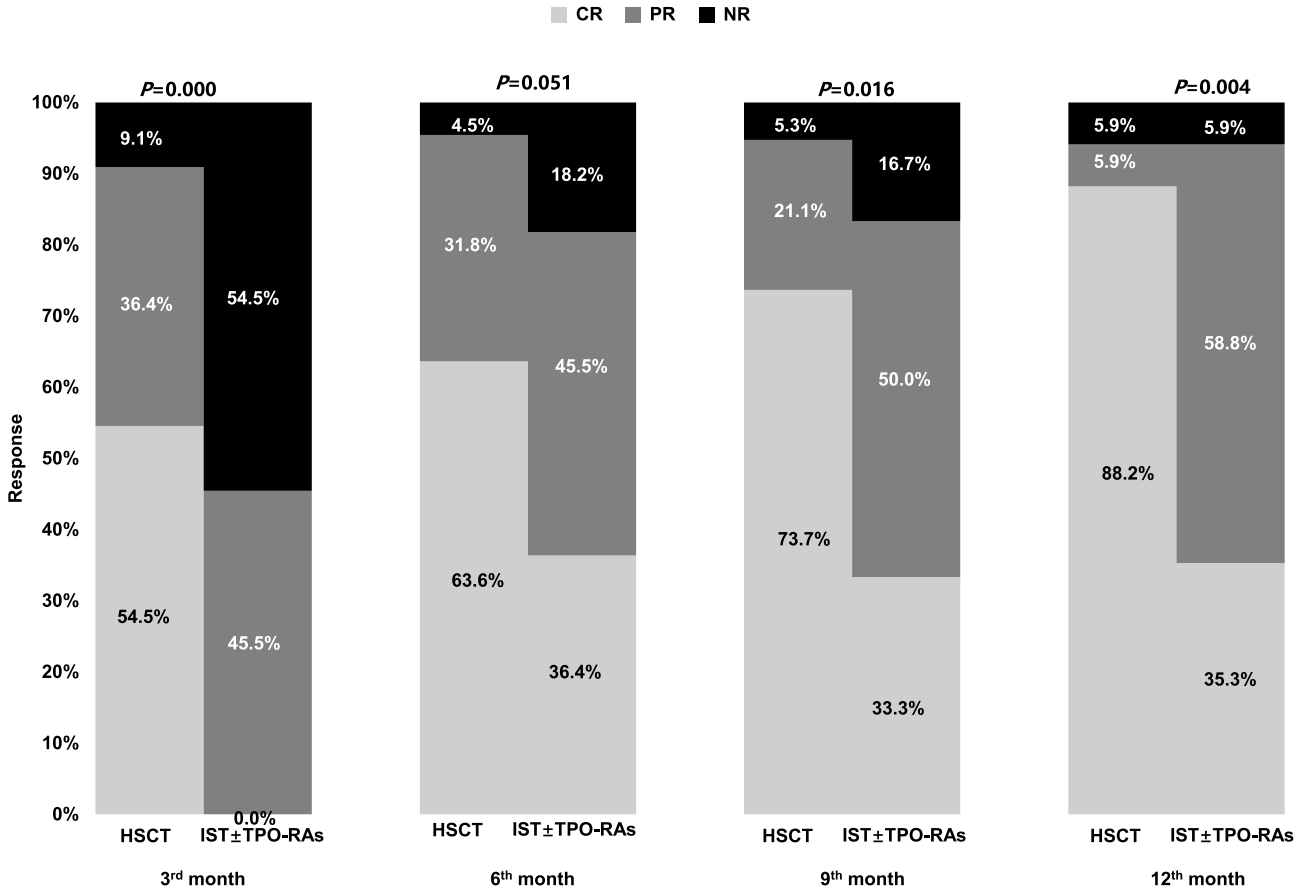

Figure 2 Response in the two regimens at the 3rd-, 6th-, 9th-, and 12th- month after treatment.

Survival Outcomes

With a median follow-up time of 19.8 (range, 0.3–38.2 months) months, the 2-year OS of all patients did not differ significantly between the HSCT (88.0%) and IST±TPO-RAs (95.7%) groups (P = 0.338; Figure 4A). We performed survival comparison between the subgroup of MSD-HSCT or haplo-HSCT and IST+TPO-RAs groups. There was a trend

Table 2 The Overall Response Between the Two Group at the Different Time

Month	Response	HSCT[%(n/N)]	IST±TPO-RAs[%(n/N)]	P value
3rd	ORR	90.9 (20/22)	45.5(10/22)	**0.001**
	NR	9.1(2/22)	54.5(12/22)	
6th	ORR	95.5 (21/22)	81.8(18/22)	0.342
	NR	4.5(1/22)	18.2(4/22)	
9th	ORR	94.7 (18/19)	83.3(15/18)	0.557
	NR	5.3(1/19)	16.7 (3/18)	
12th	ORR	94.1 (16/17)	94.1 (16/17)	1.000
	NR	5.9(1/17)	5.9(1/17)	

Notes: Two-tailed P values <0.05 were considered statistically significant. Bold values indicate statistical significance.

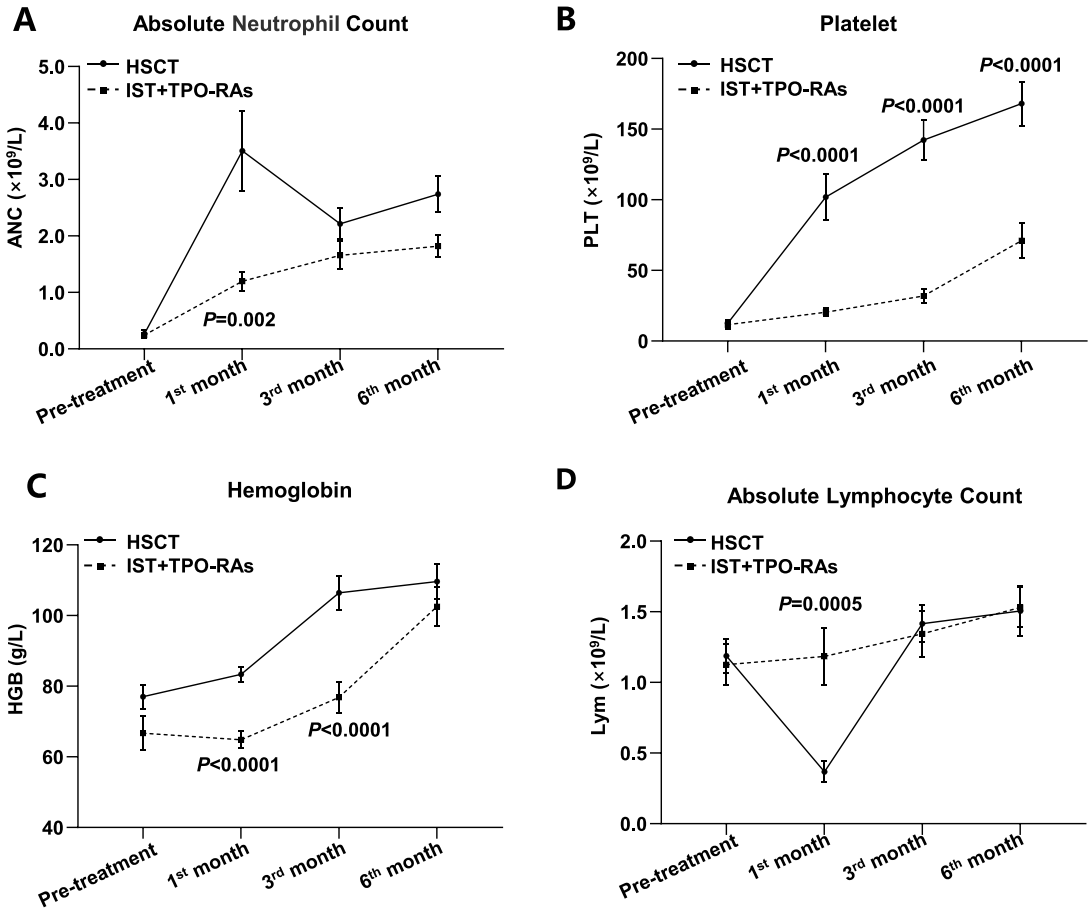

Figure 3 Blood examination changes in the two groups were observed pre-treatment and 1st-, 3rd-, and 6th-month after treatment. (**A**) HSCT has a higher absolute neutrophil count than IST±TPO-RAs group but no different at 3rd-, and 6th-month after treatment. (**B**) The HSCT group's platelets were considerably higher than those in the IST±TPO-RAs group at 1, 3, and 6 months after therapy; (**C**) The hemoglobin rises rapidly in HSCT group at 1st- and 3rd-month after treatment; (**D**) the IST±TPO-RAs group has a significant higher absolute lymphocyte counts than HSCT at 3 month after treatment.

that IST+TPO-RAs was superior to haplo-HSCT in 2-year OS (99.4% vs 77.8%), though with insignificant statistical difference (P = 0.198; Figure 4B). For patients received MSD-HSCT and IST+TPO-RAs (both n = 11) after propensity score matching (the propensity score was calculated based on a multivariate logistic regression model, which took patient age, sex, and disease status between the two groups as covariates), there was a comparable 2-year OS rate (both 90.9%, P = 0.973; Figure 4C).

The 2-year FFS were no different between total HSCT (71.4%) and IST±TPO-RAs (79.3%, P = 0.378; Figure 4D) group; or between MSD-HSCT and IST+TPO-RAs group after propensity score matching (72.7% vs 63.6%, P = 0.858; Figure 4E). The 2-year FFS were comparable between haplo-HSCT and IST+TPO-RAs (64.8% vs 73.5%, P = 0.437; Figure 4F) group even patients were much older at IST+TPO-RAs group (Supplement Table 1).

Subgroup analyses were performed on patients with SAA. The 2-year OS did not differ significantly between the HSCT (87.0%) and IST±TPO-RAs (95.5%, P = 0.317; Supplement Figure 1A) in patients with SAA. The 2-year FFS were no different between the HSCT (73.4%) and IST±TPO-RAs (78.1%, P = 0.503; Supplement Figure 1B) group.

Treatment-Related Mortality, Relapse, and Safety

One patient (4.3%) in the IST±TPO-RAs died early due to cerebral hemorrhage. Three patients (14%) in the HSCT group died within 60 days in the HSCT group for the following reasons: cerebral hemorrhage during the conditioning regimen period, circulatory failure, or sepsis. Two and 3 (10% vs 14.3%, P = 1.000) patients were relapse in the IST±TPO-RAs and HSCT group, respectively.

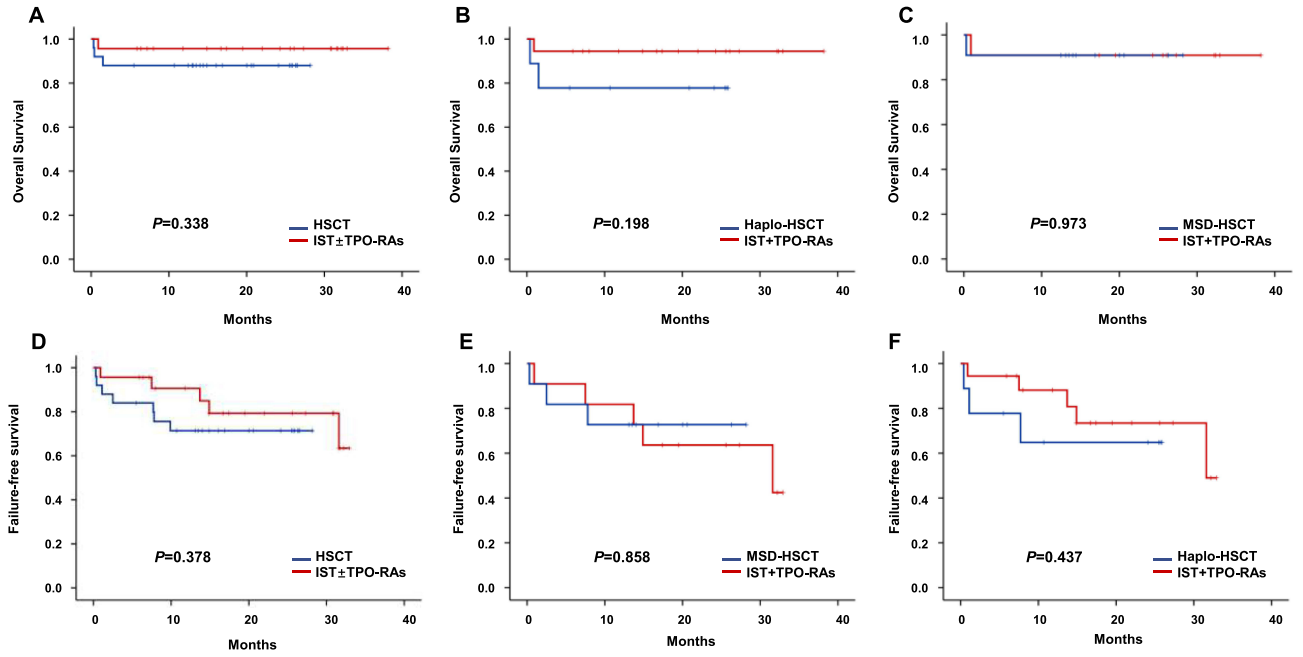

Figure 4 Failure-free survival (FFS) and overall survival (OS) in different groups. (**A**) OS and (**D**) FFS are similar in HSCT and IST±TPO-RAs groups. The OS (**C**) and FFS (**E**) were no different in MSD-HSCT (n=11) and IST+TPO-RAs (n=11) groups after propensity score matching. The OS (**B**) and FFS (**F**) in haplo-HSCT (n=9) group resemble in IST+TPO-RAs (n=18).

Of the total 23 patients in the IST±TPO-RAs group, 7 (30.4%) had serum sickness that could be relieved by steroid treatment. Seven (30.4%) patients had liver and kidney dysfunction, and 3 of them were 3 grades. Five patients (21.7%) had infection (except Epstein-Barr virus/cytomegalovirus). Edema occurred in 3 patients (all ≤grade 2). One patient experienced heart failure (grade 2).

Table 3 lists the incidence of transplant-related complications in the HSCT group. Acute GVHD occurred in 11 of the 25 patients, including 3 patients with grade I (27.3%), 7 with grade II (63.6%), and 1 with grade III (9.1%). Four patients experienced cGVHD (incidence, 16%, 4 of 25 patients had cGVHD; 2-year cumulative incidence, 25.0%). No extensively severe cGVHD were occurred and nobody died of GVHD. Fifteen patients experienced cytomegalovirus (CMV; 60.0%) and 8 patients experienced Epstein-Barr virus (EBV; 32.0%). Nine patients (36.0%) had elevated liver enzymes (4 of them were grade 3); diarrhea occurred in 7 patients (one patient was grade 4); edema occurred in 9 patients (no grade ≥3); heart failure occurred in 4 patients (3 patients were grade 3, one patient was grade 4); and 3 patients had hemorrhagic cystitis.

Table 3 Transplantation-Related Complications

Characteristics	HSCT (n=25)
CMV infection	15(60.0%)
EBV infection	8(32.0%)
Other infection	5(20.0%)
Dysfunction of liver and kidney	9(36.0%)
Edema	9(36.0%)
Diarrhea	7(28%)
Heart failure	4(16.0%)
Hemorrhagic cystitis	3(12.0%)
Acute GVHD	11(44.0%)
Chronic GVHD	4(16.0%)

Abbreviations: CMV indicates cytomegalovirus; EBV, Epstein-Barr virus; GVHD, graft versus-host disease.

Economic Analysis

The median duration of first-time hospital stays was 75 (range, 24–179) days for the IST±TPO-RAs group and 81 (range, 31–156) days for the HSCT group (P = 0.569; Table 1). The median expenses for the first-time hospitalizations were 292902 (range, 113 865–435 174) yuan in the IST±TPO-RAs group and 402756 (range, 200 116–808 954) yuan in the HSCT group (P = 0.002).

As for the IST+TPO-RAs (n = 18) and haplo-HSCT (n = 9) subgroups, the median duration of first-time hospital stays was similar (IST+TPO-RAs vs haplo-HSCT, 71 vs 104 days, P = 0.382); however, the haplo-HSCT group had significantly higher expenses compared to the IST+TPO-RAs group (IST+TPO-RAs vs haplo-HSCT = 338419 vs.487353yuan, P = 0.002, Supplement Table 1).

Discussion

AA is a bone marrow hematopoietic failure (BMF) syndrome. Its annual incidence rate in China is 0.74 per 100,000.[3] The two main treatment options for untreated SAA and TD-NSAA are allogeneic (allo-) HSCT and IST.[3,13] IST therapy use cyclosporine A (CsA) and ATG including horse-ATG (h-ATG), rabbit-ATG(r-ATG), and porcine-ATG (p-ALG). The hematological response can be accelerated and deepened more quickly and thoroughly when TPO-RAs are added to standard IST than IST alone,[7,14,15] however, MSD-HSCT can achieve longer FFS than h-/r-ATG based IST plus TPO-RAs in patients with SAA. In fact, patients without matched related donor or elder age with complication make them failed to receive HSCT. Numerous studies evaluating the safety and effectiveness of p-ALG and r-ATG in IST indicate that p-ALG exhibits comparable OS, EFS, and ORR to r-ATG at six months,[4–6] but has a better ORR at three months and lower MDS/AML transformation.[4] Since p-ALG is only available in the Chinese market and IST used p-ALG combined with TPO-RAs is rarely reported and it still remains unclear which method is better between p-ALG-based IST+TPO-RAs and HSCT, we retrospectively compared p-ALG, CsA±TPO-RAs with HSCT in patients with AA.

Studies reported that MSD-/haplo-HSCT achieved better CR rates than TPO-RAs combined with IST used r-ATG or p-ALG. Liu et al[16] compared matched related donor HSCT (MRD-HSCT) and r-ATG or p-ALG based IST+EPAG as a first choice for SAA patients at China. The MRD-HSCT group achieved faster transfusion independence, absolute neutrophil count ≥1.0 × 10^9/L (P < 0.05), as well as high percentage of normal blood routine at 6-month (86.5% vs 23.7%, P < 0.001), compared with IST + EPAG group. A multicenter retrospective study in China compared MSD-HSCT with IST+EPAG (only 4 of 47 patients used p-ALG, the remaining received r-ATG) first-line treatment of SAA in adults. Throughout the follow-up period, the CR and ORR are 15.7% and 72.6% in IST/EPAG group, respectively. Patients in the MSD-HSCT group achieved higher CR rate (79.2%) and ORR (97.9%) than IST/EPAG group (P < 0.001; P = 0.001).[8] A systematic review indirectly compared the outcomes of IST plus EPAG and haplo-HSCT in upfront treatment for SAA.[18] Ten studies were selected, including four studies used h-/r-ATG as part of IST+ EPAG and haplo-HSCT were used as first-line treatment in six studies. The ORR at 6 months was similar between the two groups (P=0.126), but the haplo-HSCT group had a significantly higher CR rate than the IST+ EPAG group (P = 0.0012). Sheng et al[19] retrospectively compared r-ATG plus CsA and HSCT in patients with TD-NSAA. The HSCT group also achieve a faster transfusion independent time than r-ATG plus CsA group (24 days vs 94 days, p < 0.001). The ORR and CR rates at 3rd, 6th, and 12th months were significant higher in HSCT (ORR, 82.1%, 78.6%, and 78.6%; CR rate, 50%, 57.1% and 60.7%) than that in r-ATG plus CsA group (ORR, 33.3%, 44.4%, and 48.1%; CR rate, 7.4%, 14.8%, and 18.5%, all P<0.05) in TD-NSAA. Our work shows that HSCT group achieved faster transfusion independence for platelets and RBCs, as well as PLT≥ 100 × 10^9/L (P < 0.05), compared with IST ±TPO-RAs group. The CR rates were higher in HSCT group than IST±TPO-RAs after treatment (Figure 2). Besides, the percentage of patients who recovered a normal blood routine was higher in the HSCT group than that in the IST±TPO-RAs (90.9% vs 54.5%, P = 0.007). However, IST ±TPO-RAs group had similar ORR with HSCT cohort after 6 months post-treatment (6th-months: 81.8% vs 95.5%, P = 0.342; 9th-months: 83.3% vs 94.7%, P = 0.557; 12th-months: both 94.1%, P = 1.000, Table 2). Our work indicated that HSCT improved the speed of hematological response than the p-ALG-based IST combined with TPO-RAs, but can achieve similar ORR after 6 months.

Studies reported that MSD-/haplo-HSCT achieved better FFS than TPO-RAs combined with IST used r-ATG or p-ALG, but the OS were no different in the two group. Huang et al[8] reported that the EFS was significantly lower in the r-ATG or p-ALG-based IST/EPAG group than in the MSD-HSCT group (71.0% vs 89.6%, $P = 0.04$), but the OS was similar between the MSD-HSCT and IST/EPAG groups (95.8% vs 96.1%, $P = 0.97$). A systematic review indirectly compared the outcomes of IST using h-/r-ATG plus EPAG and haplo-HSCT in upfront treatment for SAA concluded the same result.[18] The average 1-year as well as 2-year OS rate was similar in the IST+EPAG group and in the haplo-HSCT group (1-year OS rate: 94% and 86%, $P = 0.303$, 2-year OS rate: 89% and 84%, $P = 0.558$). The OS still no significant differences between IST+ EPAG and haplo-HSCT group after age-matched. In patients with TD-NSAA, the 5-year OS and 5-year EFS were similar between HSCT and r-ATG plus CsA group (OS: 70.3% and 56.5%, $P = 0.845$; EFS: 63.6% and 35.9%, $P = 1.445$).[19]

In our work, the 2-year OS and the 2-year FFS were similar in the IST±TPO-RAs group and HSCT group (OS: 95.7% and 88.0%, $P = 0.338$; FFS: 79.3% and 71.4%, P=0.378). In SAA subgroup, the 2-year OS and the 2-year FFS was no different in the HSCT group and IST±TPO-RA group (OS: 87.0% and 95.5%, $P = 0.317$; FFS: 73.4% and 78.1%, $P = 0.503$). Subgroup analysis between MSD-/haplo-HSCT with IST+TPO-RAs groups had similar results for long-term survivals. After propensity score matching, the 2-year OS and FFS were no different between MSD-HSCT and IST +TPO-RAs group (2-year OS: both 90.9%, $P = 0.973$; 2-year FFS: 72.7% vs 63.6%, $P = 0.858$). As for haplo-HSCT and IST+TPO-RAs cohorts, we compared baseline characteristic instead of propensity score matching in the two groups due to the small sample size. There were no different between the two groups, expect the median age in patients treated with haplo-HSCT was younger than that in IST+TPO-RAs group (Supplement Table 1). The 2-year FFS was no different in the IST+TPO-RAs and haplo-HSCT groups (both P>0.05). The estimated 2-year OS of IST+TPO-RAs was higher than haplo-HSCT group but was not statistically significant (94.4% vs 77.8%, P=0.198). Our work indicated that p-ALG-based IST±TPO-RAs could achieve similar long-term outcomes with MSD-/haplo-HSCT.

Interestingly, we found that lymphocyte count decreased in HSCT cohort and was lower than that in IST±TPO-RAs cohort at 1st-month after treatment and recovered to equivalent levels after 2 months. Previous studies of r-ATG/p-ALG-based-IST indicated more clearance of peripheral blood lymphocytes by r-ATG than by p-ALG.[4–6] Ma et al[5] retrospectively compared the efficacy and survival of untreated SAA patients who used pALG or rATG. They found that minimum number of lymphocytes was significantly lower in the r-ATG group than that in the p-ALG group at 1st-, 3rd-, and 6th-month after therapy. Deeper analysis showed that the r-ATG group had significantly lower CD4+ T cells than the p-ALG group, but there was no difference in the proportion of CD8+ T cells between the two groups. Studies found that more prolonged lymphopenia and lower lymphocyte count in r-ATG than in the h-ATG.[20] Besides, the median peak EBV copies were higher in the patients treated with r-ATG when compared with h-ATG, and alemtuzumab ($P < 0.001$).[21] A retrospective study analyzing the efficacy of p-ALG and r-ATG as pretransplant conditioning regimens for haplo-HSCT in the treatment of AA showed that the incidence of CMV and EBV infection was significantly lower in the p-ALG group than in the r-ATG group.[22] And they infer that the stronger immunosuppressive effects may possibly account for the higher incidence of CMV and EBV infection in r-ATG group than the p-ALG group. In our work, the incidence of EBV and CMV the HSCT group were 60% and 32%, respectively. Unfortunately, only a few patients in the IST±TPO-RAs had done the check. Future studies can analyze the effects of lymphocyte subsets on the incidence of EBV and CMV infection in HSCT compared with p-ALG, CsA+TPO-RAs in patients with AA.

GVHD is a common complication of HSCT. The aGVHD and cGVHD ranged at 16.7–64.0% and 8.4–39.3% for HSCT, respectively.[23–28] A meta analysis compared first-line treatment with IST and haplo-HSCT for young patients with SAA. It draw conclusion that more infections took place in haplo-HSCT group than in IST group (76% and 45%, $P < 0.01$).[29] Infections and haemorrhage were the primary reasons of death in the IST±EPAG group, whereas GVHD and infections were the primary reasons for mortality in the haplo-HSCT group.[18,29] The aGVHD and cGVHD in our work were 44.0% and 16.0%, respectively. And no extension cGVHD occurred. In the IST±TPO-RAs, the death rate was relatively low due to bleeding, while the HSCT group saw a higher death rate (albeit not statistically significant) with infections and GVHD as the most common causes of death. It should be noted that infection and GVHD may damage the quality of life, or even death.[17,30]

It is worth notice that the median expenses for the first-time hospitalizations were lower in the IST±TPO-RAs group than that in the HSCT group ($P = 0.002$), but the median duration of first-time hospital stays was no different in the two groups. Indicating that the increased medical burden may be related with higher infection and GVHD in the HSCT.

Conclusions

In conclusion, HSCT could achieved faster and higher CR rate than p-ALG-based IST±TPO-RAs in patients with AA. Compared with HSCT, p-ALG-based IST±TPO-RAs has lower ORR at three months but achieves similar outcomes of 6^{th}-month ORR, 2-year FFS and 2-year OS with HSCT, especially in elder patients. Moreover, p-ALG-based IST±TPO-RAs has the advantage of significantly lower first-time hospitalizations cost. Thus, p-ALG-based IST±TPO-RAs is an alternative treatment for patients who were not candidates for transplant. AA is a rare hematologic disorder. Although our sample size is insufficient, we directly compared the role of p-ALG in combination with CsA and TPO-RAs with HSCT in AA. However, this is a single-center, retrospective study, and it is needs to be confirmed by multicenter prospective study.

Data Sharing Statement

Data are available on request from the corresponding author.

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Author Contributions

All authors made a significant contribution to the work reported, whether that is in the conception, study design, execution, acquisition of data, analysis, and interpretation, or in all these areas; took part in drafting, revising, or critically reviewing the article; gave final approval of the version to be published; have agreed on the journal to which the article has been submitted; and agree to be accountable for all aspects of the work.

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Disclosure

The authors declare no competing interests in this work.

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