The use of splenectomy to manage platelet transfusion refractoriness due to anti-human leukocyte antibodies in allogeneic stem cell transplantation

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Abstract

In patients undergoing hematopoietic stem cell transplantation (HSCT), refractoriness to platelet transfusion has been associated with graft failure, delayed engraftment, early mortality and decreased overall survival. Therapeutic strategies include plasma exchange, immunoglobulins, rituximab, and splenectomy. We describe here three patients with refractoriness to platelet transfusion due to anti-human leukocyte antibodies who were splenectomized before HSCT (two cases) and after HSCT (one case) due to the lack of efficacy of other therapies. Splenectomy was uneventful. All three patients achieved a full donor engraftment. We suggest that splenectomy is feasible and effective in HSCT patients to reduce the risk of graft failure or delayed engraftment.

Introduction

Immune-mediated refractoriness to platelet transfusions is observed in patients who receive non-leukodepleted hemocomponents and develop anti-HLA antibodies.[1,2] This complication may have a negative impact on the success of a hematopoietic stem cell transplant (HSCT) because it has been related to graft failure.[3-7] Moreover, the presence of anti-HLA antibodies has been associated with a delay in the time of neutrophil and platelet engraftment, early mortality and decreased long-term overall survival.[5] The presence of a spleen and the number of anti-HLA antibodies were the factors that predict the yield of platelet transfusion in HSCT patients.[8]

Therapeutic strategies for platelet transfusion refractoriness include removing the existing antibodies by plasma exchange, inhibiting antibody activity by immunoglobulin, and blocking B cells or plasma cells by rituximab.[9] Splenectomy can improve the yield of platelet transfusion although it does not alter the number and type of anti-HLA antibodies.[8]

We describe three patients who developed refractoriness to platelet transfusion due to anti-HLA antibodies before HSCT who were successfully managed by splenectomy. Table 1 shows the main demographic and clinical characteristics of the three patients.

Case Report #1

K.L. is a 3-year-old female from Georgia diagnosed with myelodysplastic syndrome who was treated mainly with supportive prophylactic non-leukodepleted platelet and red cell transfusions for 19 months until she was referred to our center for an allogeneic HSCT. At admission, she presented with refractoriness to platelet transfusion. The search of anti-HLA antibodies showed the presence of anti-HLA class I (ELISA PakAuto and Quick Screen; GTI Diagnostics, Brookfield, WI, USA). The patient was treated initially with two courses of intravenous immunoglobulins (0.8 g/kg) followed by four courses of dexamethasone (4×40 mg/day) and rituximab (4×375 mg/m^2).[10] These therapies did not result in significant increment of post-transfusion platelet count so, after three months, a laparoscopic splenectomy was decided upon, preceded by vaccination for capsulated bacteria (one dose of 23-valent pneumococcal polysaccharide vaccine, one dose of 4-valent polysaccharide meningococcal vaccine and one dose of hemophilus B conjugate vaccine). Splenectomy was uneventful and the patient was discharged after 8 days. The platelet count increased from 12×10^9/L pre-surgery to 51×10^9/L two days after splenectomy. The median transfusion-dependency dropped from three transfusions/week before surgery to none after splenectomy. Five months after splenectomy, the patient underwent an unmanipulated haploidentical stem cell transplant. A stable neutrophil count >0.5×10^9/L and a platelet count >50×10^9/L were achieved after 16 and 42 days from transplant, respectively. The patient was discharged on day + 19 post-HSCT. After 11 months from HSCT the patient is well (Lansky play score 100%), in amoxicillin prophylaxis and in complete hematological remission with blood count always within the normal range and full donor chimerism. The maximum count of platelet reached after splenectomy was 342×10^9/L.

Case Report #2

T.Z. is an 8-year-old male from Georgia diagnosed with myelodysplastic syndrome and treated supportively for 8 months with non-leukodepleted platelet and red cell transfusions until he was referred to our center for allogeneic HSCT. At admission, the patient had refractoriness to platelet transfusion and the search for anti-HLA class I antibodies was positive (ELISA PakPlus; Gen Probe Diagnostics, San Diego, CA, USA). The patient was treated with three courses of intravenous immunoglobulins (0.8 g/kg), followed by four courses of dexamethasone (4×40 mg/day) and rituximab (4×75 mg/m^2).[10] After 4 months, due to the persistence of a high dependency to platelet transfusions, the patient underwent a laparoscopic splenectomy preceded by vaccination for capsulated bacteria (one dose of 23-valent pneumococcal polysaccharide vaccine, one dose of 4-valent polysaccharide meningococcal vaccine and one dose of hemophilus B conjugate vaccine). The operation was uneventful and the patient was discharged after 9 days. The platelet count was 4×10^9/L pre-surgery and increased to 19×10^9/L seven days after splenectomy. Before surgery, the median requirement was two-platelet transfusions/week, whereas after splenectomy the patient required no further transfusions. One month after splenectomy, the patient underwent a haploidentical stem cell transplant. Neutrophil and platelet engraftment occurred 14 days and 27 days, respectively and the patient was discharged on day + 27 post-HSCT. At the last follow-up, at + 18 months after

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Key words: anti-HLA; splenectomy; alloimmunized patients; transfusion refractoriness; allogeneic stem cell transplantation.

Contributions: SC and MM wrote the manuscript, MM and AP collected the data, FC performed splenectomies, PP performed the tests for anti-HLA antibodies; SC, MDB, RB took care of the patients. All authors approved the manuscript.

Conflict of interest: the authors declare no potential conflict of interest.

Received for publication: 24 August 2015.
Revision received: 19 December 2015.
Accepted for publication: 29 December 2015.

Pediatric Reports 2016; 8:6159
doi:10.4081/pr.2016.6159

HSCT, the patient is well, active (Lansky play score 90%), in amoxicillin prophylaxis and in complete hematological remission with blood count always within the normal range and full donor chimerism. The maximum count of platelet reached after splenectomy was 491×10⁹/L.

Case Report #3

V.B. is a 6-year-old male from Ukraine, diagnosed with Juvenile Myelomonocytic Leukemia (JMML) who was treated with hydroxyurea, four courses of cytarabine, and supportive care including non-leukodepleted transfusions for 8 months until he was referred to our center for an unrelated HSCT. Before HSCT, the patient presented with refractoriness to platelet transfusions that was initially attributed to his underlying disease with splenomegaly (+ 4 cm below left costal margin), but subsequently it was considered the result of allo-immunization because the search for HLA antibodies class I and anti-GPIb/IX, GPIa/IIa and GP IIb/IIIa (ELISA Pak 12; GTI Diagnostics, Brookfield, WI, USA) resulted positive. Because the HSCT was already scheduled it was decided to proceed, modifying the conditioning regimen by adding a course of plasma exchange, a course of rituximab (1×375 mg/m²) with high dose intravenous methylprednisolone (500 mg/m²/week) and splenic irradiation. Neutrophil engraftment occurred at day +22 after transplant whereas platelet engraftment did not occur and the patient remained transfusion-dependent with a median requirement of four transfusions /week. Given the persistence of splenomegaly, on day + 91 post-HSCT, a laparoscopic splenectomy was carried out. The surgery was uneventful and the patient was discharged after three weeks. Thirty-five days after splenectomy, the platelet count was 186×10⁹/L with a maximum of 428×10⁹/L two months later. At 10 months post-HSCT, the patient is well (Lansky play score 100%), in amoxicillin prophylaxis and in complete hematological remission with blood count always within the normal range and full donor chimerism.

Discussion

Platelet refractoriness may occur through a non-alloimmune or an alloimmune mechanism, the latter being caused by sensitization to foreign Class I HLA and, less frequently, to human platelet polymorphic antigens (HPA).[11] This complication has been reported in between 7-34% of patients with hematological or oncological diseases, especially in patients who do not receive leuko-depleted or irradiated hemoproducts.[11,12] In a randomized trial, the use of leukodepletion or ultraviolet B irradiation of platelets decreased alloimmunization from 13% to 3% and 5%, respectively.[13] All three patients described here came from centers where leukodepletion and/or irradiation of blood products were not routinely performed.

The management of platelet refractoriness is based on the modification of the type of platelet product administered or on the modification of the patient immune response. In the first case, the selection of an HLA-matched donor gives the highest post-transfusion platelet increment but it requires the availability of a large pool of dedicated HLA-typed donors. The process of donor selection is time-consuming without the certainty of finding a matched donor; this procedure is also not ideal for urgent management of bleeding. Alternatively, a platelet cross-matching test with patient serum can be used to more rapidly identify the best platelet unit available.[14] In our center, both procedures were not feasible because of an inadequate size of donor pool and the inability to perform HLA screening on every donor, so that the only feasible procedure to improve the platelet transfusion yield was to use platelet units derived from a pool of donors. However, this policy did not result in a significant reduction of transfusion frequency, which remained on a daily or two-daily basis. The modification of immune response by high-dose of steroids, immunoglobulins and rituximab was not associated with any improvement of patient transfusion-dependency. Considering that HLA sensitization can be a major obstacle to successful engraftment in HSCT,[6] and may lengthen the period of transfusion-dependency, increasing the risk of severe bleeding episodes,[15] we opted for splenectomy. Despite the low number of platelets at surgery, the laparoscopic procedure was well tolerated without any significant bleeding. This allowed the first two patients to undergo myeloablative chemotherapy and to engraft both for neutrophils and platelets with-

Table 1. Main demographic, clinical, and transplant characteristics of three patients.

	First case	Second case	Third case
Gender	Female	Male	Male
Country of origin	Georgia	Georgia	Ukraine
Underlying disease	Myelodysplasia	Myelodysplasia	JMML
Age at diagnosis, years	3	8	6
Type of donor	Haploidentical mother	Haploidentical mother	HLA matched unrelated donor (10/10)
Myeloablative conditioning regimen	BU 3×3.2 mg/kg/day; TT 1×5 mg/kg/day; Flu 3×50 mg/m²/day	TBI 2 Gy; BU 3×3.2 mg/kg/day; TT 2×5 mg/kg/day; Flu 3×50 mg/m²/day	RT 2 Gy (on spleen); BU 4 3.8 mg/kg/day; ATG 3×3.75 mg/kg/day; Cy 2×60 mg/kg/day; Mel 1×140 mg/m²/day
GVHD prophylaxis	Tacrolimus/cyclosporine; Cy 2×50 mg/kg/day (day + 3 and +4); MMF	Tacrolimus/ cyclosporine; Cy 2×50 mg/kg/day (day + 3 and +4); MMF	Intravenous cyclosporin 3 mg kg/day; MTX 15 mg/m² day + 1; MTX 10 mg/m² day + 3, +6, +11
Number of infused CD34+ cells	4.68×10⁶/kg	5.32×10⁶/kg	4.16×10⁶/kg
Neutrophil engraftment (>0.5×10⁹/L)	+ 16 days	+ 14 days	+ 22 days
Platelet engraftment (>50×109/L)	+ 42 days	+ 27 days	+ 105 days after splenectomy
Blood count at last follow up	+ 11 months; Hb 12.9 g/dL; PLT 319×10⁹/L; WBC 11.8×10⁹/L (neutrophils 3.1×10⁹/L)	+ 18 months; Hb 14.7 g/dL; PLT 500×10⁹/L; WBC 13.2×10⁹/L (neutrophils 4.3×10⁹/L)	+ 10 months; Hb 14.2/g/dL; PLT 428×10⁹/L; WBC 10.6×10⁹/L (neutrophils 4×10⁹/L)

JMML, juvenile myelomonocytic leukemia; TBI, total body irradiation; RT, radiation therapy; BU, busulfan; TT, thiotepa; Mel, melphalan; Flu, fludarabine; ATG, anti-thymocyte globulin; Cy, cyclophosphamide; MTX, metothexate; MMF, mycophenolate mofetil.

out any significant delay in the first two cases described. Interestingly enough, the third alloimmunized patient did not engraft normally for platelets, despite the modification of the conditioning regimen with the addition of plasmapheresis, dexamethasone, rituximab and splenic radiotherapy, and continued to be platelet transfusion-dependent until splenectomy was performed at three months after transplant. Marktel *et al.* showed that myeloablative and immunosuppressive conditioning before HSCT was not able to modify the number and specificities of anti-HLA antibodies in eight alloimmunized patients who rejected their graft.[8] On the contrary, in a thalassemic alloimmunized patient who had a secondary graft failure, a splenectomy before the second HSCT did result in higher platelet transfusion increment than the first HSCT (P=0.03). Splenectomy, though not altering the anti-HLA antibody production, can limit its negative impact on the life of both exogenous and endogenous platelets. This finding is in line with the data reported recently by Palandri *et al.*[16] who showed that in patients with steroid-refractory immune thrombocytopenia, splenectomy had a response rate of 88.5% after a median follow-up of 20 years; moreover, the response rate was similar to or even higher than rituximab in patients with non-splenic uptake of platelets.

Conclusions

In conclusion, in alloimmunized patients, laparoscopic splenectomy is a valuable option before HSCT to prevent graft failure, speed platelet engraftment and reduce transfusional burden after HSCT in patients with anti-HLA antibodies. Further studies and longer follow-up is needed to assess the impact of this choice on long-term infectious morbidity.

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