

Successful allogeneic bone marrow transplantation using immunosuppressive conditioning regimen for a patient with red blood cell transfusion-dependent pyruvate kinase deficiency anemia

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Abstract

Pyruvate kinase deficiency (PKD) is the rare glycolytic enzyme defect causing hemolytic anemia. Treatments are mainly red cell transfusion and/or splenectomy, leading to iron overload. Allogeneic bone marrow transplantation (BMT) is alternatively curative treatment for severe PKD. The intensity of conditioning is often controversial because of higher risk of graft failure and organ damage. Here, we present a transfusion-dependent PKD patient undergoing BMT from an HLA-identical sibling using intensively immunosuppressive conditioning regimen. This report suggests that BMT using immunosuppressive conditioning regimen may be a feasible and effective treatment for patients with severe PKD with iron overload. We suggest the timing of the transplantation at an earlier age in severe PKD predicted from gene mutation is preferred before cumulative damage of transfusion.

Introduction

Red cell pyruvate kinase deficiency (PKD) is a common glycolytic defect that causes congenital non-spherocytic chronic hemolytic anemia. The hemolysis severity in PKD patients is highly variable.¹ PKD treatment mainly involves supportive care with red blood cell (RBC) transfusion and splenectomy. PKD is amenable to gene therapy, but there are no clinical trials using gene therapy or gene editing.² Mitapivat is, an allosteric activator of PK in red cells, effective for some patients with at least one missense mutation.³ In patients undergoing frequent transfusion, organ damage due to iron overload is a major concern. The only

curative therapy for severe PKD is hematopoietic stem cell transplantation (HSCT). However, iron overload is associated with poor prognosis in patients undergoing HSCT.⁴ In hemoglobinopathies, HSCT following myeloablative conditioning (MAC) is related to a higher incidence of treatment-related mortality (TRM); moreover, reduced intensity conditioning (RIC) is also related to graft failure. Graft failure in hemoglobinopathies may be due to significant marrow hyperplasia and allosensitization caused by repeated blood transfusions.⁵⁻⁷ However, few reports and limited data are available regarding bone marrow transplantation (BMT) following other than MAC for PKD. We report successful BMT following immunosuppressive conditioning regimen in a transfusion-dependent severe PKD patient (> 30 years old) with organ damage caused by iron overload.

Case Report

The patient was a 32-year-old Japanese man who was referred to us for HSCT because of severe anemia. He was born with severe hemolytic anemia and jaundice. He was diagnosed with PKD based on the PK activity in the RBCs. He had been receiving frequent RBC transfusions since his birth. He underwent splenectomy at the age of 5. However, his anemia did not subside. Thereafter, frequent RBC transfusion for maintaining the hemoglobin level at 7.0 g/dL with iron chelation was continued for several years. He also underwent cholecystectomy because of hemolysis-related gallstone complication at the age of 19 years. He was diagnosed with severe PKD at the age of 32 years with mutations of the *PKLR* gene (Exon5 c.434 C del: PK Beppu).⁸ He had severe anemia and had undergone frequent RBC transfusion, therefore, he was unable to obtain full-time employment and had an impaired quality of life. Therefore, he wanted to undergo BMT as curative therapy. Prior to BMT, his serum ferritin level was 740 ng/mL. Abdominal magnetic resonance imaging (MRI) T2 sequences showed liver iron burden (Figure 1). Although the MRI did not reveal cardiac iron burden, we predicted potential organ damage as hepatic hemosiderosis. He also had broad, high-titer and non-specific anti-human leukocyte antigen (HLA) class I antibody. Although their antibody had the capability of exerting adverse effects on engraftment, we administered Rituximab 375 mg/m² thrice weekly and plasma exchange before BMT.

He underwent BMT with the

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Contributions: MS designed the work, and wrote the initial draft of the manuscript. TD, doctor following this case, assisted in the preparation of the manuscript. All other authors have contributed to treatment of this case, and critically reviewed the manuscript. All authors approved the final version of the manuscript, and agree to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. MK gave final approval of the version to be published.

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immunosuppressive conditioning regimen from his HLA identical sister at the age of 32 (Figure 2). Conditioning regimens included 3.6 Gy total body irradiation (day -8), fludarabine 30 mg/m² (×6 days, from day -7

to -4), melphalan 90 mg/m² (×2 days, day -3 and day -2), and rabbit antithymocyte globulin 1.25 mg/kg (×4 days, day -7 to -4). Graft-versus-host disease (GVHD) prophylaxis included tacrolimus and short methotrexate. The number of infused nucleated cells was 2.5 × 10⁸ cells/kg and that of CD34+ cells was 1.7×10⁶ cells/kg. The early post-transplantation period was uneventful. Stable neutrophil engraftment occurred on day 17. The last blood and platelet transfusion days were day 22 and day 23. The patient presented with fever on day 19. Fever associated with engraftment was successfully treated with methylprednisolone. On day 55, he presented with a rash on <25% of the body surface area; however, he did not have diarrhea and icterus. He was diagnosed with grade 1 GVHD. At the time of discharge, he had mild chronic skin GVHD but was in a state of complete donor chimerism. Three years following the BMT, the patient's hemoglobin level was 15 g/dL without any other complications, and he was able to work a regular job. His health-related Quality of life (QOL) was assessed using the Japanese version of the SF-36.^{9,10} His SF-36 scores of physical function (PF) increased from 75 points before BMT to 100 points after BMT. Role physical (RP) increased from 81 points to 100 points, general health (GH) increased from 47 points to 70 points, and vitality (VT) increased 75 points to 93.8 points, respectively. Now he can take part in a marathon for interest, despite he was barely able to walk a little way before BMT.

Mitapivat was associated with a rapid increase in the hemoglobin level in 50% of adults with PKD.³ However, some patients with non-missense mutations have no significant effect.³

Iron overload is an adverse prognostic factor related to poor overall survival (OS) and graft versus host disease (GVHD) in patients undergoing HSCT.^{4,12} The threshold for serum ferritin level is set at 1,000 ng/mL

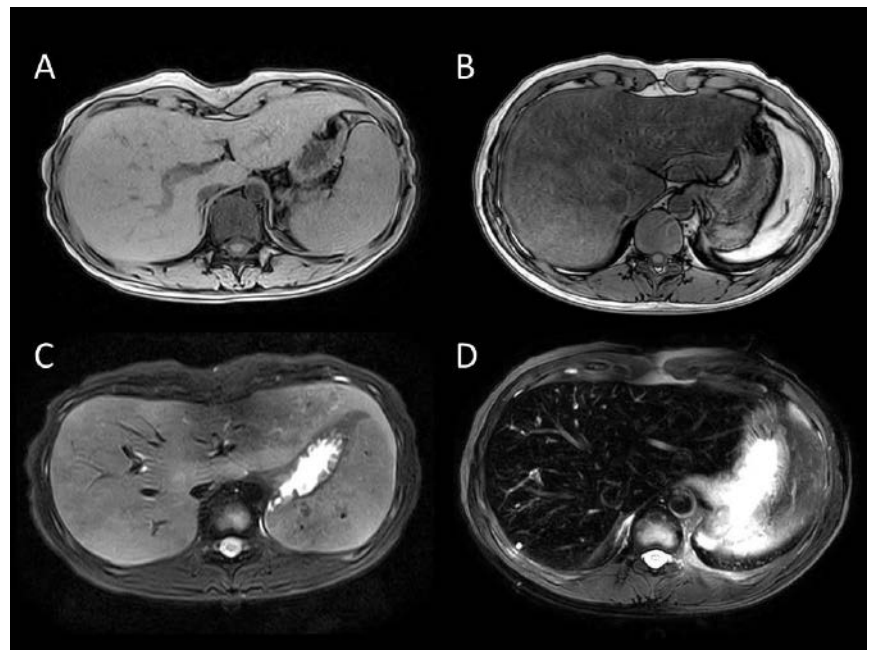


Figure 1. Abdominal MRI T1 (A,B) and T2(C,D) sequences A and C are healthy control livers. B and D are patient's livers. Both abdominal MRI T1 and T2 sequences show hypointense liver due to iron deposition in patient (B,D).

Discussion and Conclusions

We have described the successful management of a patient with severe PKD treated with BMT using the immunosuppressive conditioning regimen because of liver iron overload. In thalassemia patients, BMT resulted in higher Health related QOL in both physical and mental aspects compared to blood transfusion plus iron chelation.¹¹ BMT also resulted in dramatically improved quality of life in our patient. But the stratification of PKD severity is unclear; therefore, it is difficult to assess the indication for transplantation. Recently, remarkable investigational therapies include gene therapy and Mitapivat. Monogenic disorders such as PKD are potentially amenable to gene therapy. There are no clinical trials using gene therapy or gene editing, but attempts to correct PK deficiency in mouse models using lentiviral vectors have been reported.² Mitapivat is an oral, small-molecule allosteric activator of PK in red cells.

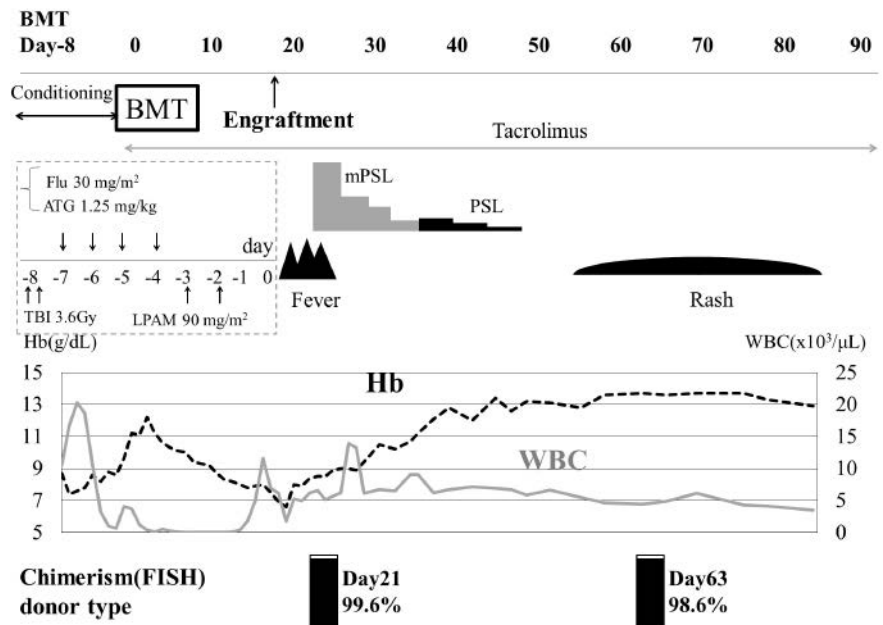


Figure 2. Clinical course after transplantation. Stable neutrophil engraftment occurred on day 17. Although he was diagnosed with grade 1 GVHD, he had been in a state of complete donor chimerism and there were no major complications during the course. mPSL: Methylprednisolone, Flu: Fludarabine, ATG: Rabbit antithymocyte globulin, LPAM: Melphalan

for the detection of iron overload.¹³ However, Brittenham *et al.* reported that serum ferritin levels had a poor correlation with liver iron concentration (LIC) using MRI in pediatric patients.^{14,15} In our patient, the pre-transplant ferritin level was 740 ng/mL; however, abdominal MRI T2 sequences revealed liver iron burden. Recently, these data clearly show that iron overload is common in PKD patients regardless of transfusion such as thalassemia patients.¹⁶ MRI should be considered routinely regardless of transfusion. Van Straaten *et al.* reported 16 cases of PKD treated with HSCT between 1996 and 2015.¹⁷ Although the intensity of conditioning was diverse, the 3-year OS rate was 65%, and the only poor prognostic factor was age > 10 years. The pre-transplant ferritin level was not a factor in this cohort. It is noteworthy that the rate of grade 3-4 GVHD and the mortality attributable to GVHD were relatively high. The underlying reason remains unclear; however, detailed studies are needed. In hemoglobinopathies, the rate of graft rejection is 10-30%.¹⁸ In adult patients with thalassemia, their non-rejection mortality was also higher than that of younger patients owing to organ damage.¹⁹ It may be possible that adult PKD patients require reduction in the conditioning regimen for organ complication; however, a certain degree of immunosuppressive conditioning regimen is necessary for the eradication of the massive erythroid hyperplasia, such as thalassemia. The intensity of our conditioning regimen classified as neither RIC nor MAC, we called immunosuppressive regimen, may be appropriate for PKD with organ damage.

The homozygous *PKLR* mutations (Exon 5 c.434 C del: PK Beppu) in our patient have been previously described by Kanno *et al.*⁸ PK Beppu associated severe PKD in Japanese.⁸

PK Beppu is frameshift mutation due to one base deletion. Therefore, Mitapivat may be not effective for our patient. The recent data revealed significantly better survival in PKD patients who underwent transplantation before the age of 10 years.¹⁷ In patients with PKD, splenectomy was associated with a decreased transfusion burden in 90% of patients,²⁰ but the effect was insufficient for our patient. Consideration of HSCT at an earlier age in severe PKD predicted from gene mutation and limited effect of

splenectomy like our patient may be warranted before cumulative damage of transfusion. Although further research is required on the intensity of the regimen and appropriate timing of HSCT, our experience demonstrates that BMT from matched sibling donors using immunosuppressive conditioning regimen can be a curative therapeutic option in severe PKD patients with organ damage.

References

1. Warang P, Kendar P, Ghosh K, Colah R. Molecular and clinical heterogeneity in pyruvate kinase deficiency in india. *Blood cells Mol Dis* 2013;51:133-7.
2. Garcia-Gomez M, Calabria A, Garcia-Bravo M, et al. Safe and Efficient Gene Therapy for Pyruvate Kinase Deficiency. *Mol Ther* 2016;24:1187-98.
3. Grace RF, Rose C, Layton DM, et al. Safety and Efficacy of Mitapivat in Pyruvate Kinase Deficiency. *N Engl J Med* 2019;381:933-44.
4. Pullarket V, Blanchard S, Tegtmeier B, et al. Iron overload adversely affects outcome of allogeneic hematopoietic cell transplantation. *Bone Marrow Transplant* 2008;42:799-805.
5. Walters M, Hardy K, Edwards S, et al. Pulmonary, gonadal, and central nervous system status after bone marrow transplantation for sickle cell disease. *Biol Blood Marrow Transplant* 2010;16:263-72.
6. Jannone R, Casella J, Fuchs E, et al. Results of minimally toxic nonmyeloablative transplantation in patients with sickle cell anemia and beta-thalassemia. *Biol Blood Marrow Transplant* 2003;9:519-28.
7. Lucarelli G, Gaziev J. Advances in the allogeneic transplantation for thalassemia. *Blood Rev* 2008;22:53-63.
8. Baronciani L, Bianchi P, Zanella A. Hematologically important mutations: red cell pyruvate kinase. *Blood Cells Mol Dis* 1996;22:85-9.
9. Fukuhara S, Bito S, Green J, et al. Translation, adaptation, and validation of the SF-36 Health Survey for use in Japan. *J Clin Epidemiol* 1998;51:1037-44.
10. Fukuhara S, Ware JE, Kosinski M, et al. Psychometric and clinical tests of

validity of the Japanese SF-36 Health Survey. *J Clin Epidemiol* 1998;51:1045-53.

11. Javanbakht M, Keshtkaran A, Shabaninejad H, et al. Comparison of Blood Transfusion Plus Chelation Therapy and Bone Marrow Transplantation in Patients with β -Thalassemia: Application of SF-36, EQ-5D, and Visual Analogue Scale Measures. *Int J Health Policy Manag* 2015;4:733-40.
12. Atilla E, Topark SK, Demirel T. Current review of iron overload and related complications in hematopoietic stem cell transplantation. *Turk J Hematol* 2017; 34:1-9
13. Knovich MA, Storey JA, Coffman LG, et al. Ferritin for the clinician. *Blood Rev* 2009;23:95-104.
14. Brittenham GM, Cohen AR, McLaren CE, et al. Hepatic iron stores and plasma ferritin concentration in patients with sickle cell anemia and thalassemia major. *Am J Hematol* 1993;42:81-5.
15. Majhail NS, Defor T, Lazarus HM, Burns LJ. High prevalence of iron overload in adult allogeneic hematopoietic cell transplant survivors. *Biol Blood Marrow Transplant* 2008;14:790-4.
16. van Beers EJ, van Straaten S, Morton DH, et al. Prevalence and management of iron overload in pyruvate kinase deficiency: report from the Pyruvate Kinase Deficiency Natural History Study. *Hematologica* 2019;104:51-3.
17. van Straaten S, Bierings M, Bianchi P, et al. Worldwide study of hematopoietic allogeneic stem cell transplantation in pyruvate kinase deficiency. *Haematologica* 2018;103:82-6.
18. Lucarelli G, Andreani M, Anelucci E. The cure of thalassemia by bone marrow transplantation. *Blood Rev* 2002;16:81-5.
19. Gaziev J, Sodani P, Polchi P, et al. Bone marrow transplantation in adults with thalassemia: treatment and long term follow up. *Ann N Y Acad Sci* 2005;1054:196-205.
20. Grace RF, Bianchi P, van Beers EJ, et al. Clinical spectrum of pyruvate kinase deficiency: data from the Pyruvate Kinase Deficiency Natural History Study. *Blood* 2018;131:2183-92.