



ARTÍCULO ORIGINAL

Second allogeneic peripheral blood stem cell transplants with reduced-intensity conditioning

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ABSTRACT

In two institutions in México, twelve patients were given a second allogeneic stem cell transplantation, using the "Mexican" non-myeloablative preparative regimen. Eight had a malignant condition (six acute leukemias, one myelofibrosis and one myelodysplasia), eleven individuals were allografted twice from the same donor and in one case, cells from two different umbilical cords were used. The median time to conduct the second allograft after the first one was 6 months (range 1-41). The five patients who failed to engraft after the first transplant failed also to engraft after the second one; all of them had been heavily transfused. Only three patients were successfully rescued with the second transplant, two with acute leukemia and one with aplastic anemia. Seven patients are alive 10-41 months (median 35) after the second transplant, but only three (25%) remain disease-free. The 52-month overall survival (SV) of the patients is 58%, whereas the median overall SV has not been reached, being above 52 months. Conducting a second allograft may be useful to rescue some individuals relapsing after a first hematopoietic allotransplant.

Key words. Allograft. Second. Failure. Relapse. Non-ablative.

INTRODUCTION

Allogeneic stem cell transplantation is widely used in the treatment of several malignant and non-malignant conditions; it is associated with a relatively low risk of disease recurrence.¹ Second allogeneic stem cell transplantation (2-SCT) may be effective salvage therapy in some patients, after an initial either auto or allogeneic transplant;²⁻⁴ 2-SCT are conducted as a result of either relapse of the disease² or graft failure.³⁻⁴ Conventional transplant conditioning has a relatively high early trans-

Segundos trasplantes hematopoyéticos con esquemas de acondicionamiento de intensidad reducida

RESUMEN

En dos instituciones en México se llevaron a cabo doce segundos trasplantes de células hematopoyéticas usando el "método mexicano" de acondicionamiento no mieloablativo. Ocho pacientes tenían una enfermedad maligna (seis leucemias agudas, una mielofibrosis y una mielodisplasia). Once sujetos se retrasplantaron del mismo donador y en un caso se emplearon células hematopoyéticas de dos diferentes cordones umbilicales. La mediana del tiempo transcurrido entre los dos trasplantes fue de seis meses (rango 1 a 41). Los cinco pacientes que no se injertaron con el primer trasplante tampoco se injertaron con el segundo; todos ellos habían sido multitransfundidos antes de los trasplantes. Sólo tres pacientes se pudieron rescatar con el segundo trasplante, dos con leucemia aguda y uno con anemia aplásica. Siete pacientes están vivos 10 a 41 meses (mediana 35) después del segundo trasplante, pero sólo tres (25%) se encuentran libres de enfermedad. La supervivencia (SV) global a 52 meses es de 58%, en tanto que la mediana de SV no se ha alcanzado y es mayor de 52 meses. Hacer un segundo trasplante hematopoyético puede rescatar a algunos pacientes quienes recaen después de un trasplante de médula ósea.

Palabras clave. Injerto. Segundo. Falla. Recidiva. No-ablativo.

plant-related mortality; however, in recent years, several groups of investigators have developed reduced-intensity conditioning (RIC) regimens, which lead to engraftment of donor lymphoid cells and hematopoietic stem cells without the extra-hematologic toxicities of traditional myeloablative transplants.⁴⁻¹⁵ We analyze herein the features of a group of 12 individuals who were allografted with a RIC regimen and who received later on a second allograft, employing also a RIC, because of relapse of the disease (7 patients) or as the result of failure of the initial allograft (5 patients).

MATERIAL AND METHODS

Patients

All the patients given consecutively a second allogeneic peripheral blood stem cell transplantation in the Centro de Hematología y Medicina Interna de Puebla (Puebla, México) and in the Hospital Universitario de Nuevo León (Monterrey, México) since 1993 were accrued in the study. Informed consent to perform the grafts was obtained from all individuals and their donors.

Allografting procedure

For both the first and second allografts, the "Mexican" non-myeloablative conditioning regimen to conduct allografts was used as previously described.⁴⁻¹⁵ Briefly, filgrastim (10 µg/kg/day) was delivered to the donors, in all cases HLA-identical siblings, on days - 5 to + 2. The apheresis procedures were performed on days 0, + 1 and + 2 by means of a Haemonetics V-50 PLUS machine (Haemonetics Corporation, Braintree MA) or a Baxter C-3000 PLUS machine (Baxter Healthcare, Deerfield IL),¹⁶ and immediately infused to recipients. Conditioning on the recipient consisted in oral busulphan, 4 mg/kg delivered on days - 6 and - 5; i.v. cyclophosphamide, 350 mg/m² on days - 4, - 3 and - 2; i.v. fludarabine, 30 mg/m² on days -4, -3 and -2; oral cyclosporin A (CyA) 5 mg/kg was started on day - 1 and i.v. methotrexate 5 mg/m² was delivered on days + 1, + 3, + 5 and + 11. Oral CyA was continued through day 180, with adjustments according to serum CyA levels; it was then tapered along 30-60 days; in the case of 2-SCT, the immunosuppression was delivered along 30 days instead of the 180. Ondansetron (1 mg intravenous every hour during 4 h after chemotherapy), ciprofloxacin (250 mg *bid*) and itraconazole (100 mg *bid*) were used in all patients; antibiotics

and antimycotics were used until more than 500 granulocytes / ul were present. The products of the PBSC apheresis were infused as outpatients on days 0, 1 and 2.

Apheresis product studies

Enumeration of the total white blood, mononuclear (MNC) and CD34 positive cells was done by flow-cytometry¹⁷ in an EPICS Elite ESP apparatus (Coulter Electronics, Hialeah, FL), using the HPCA-2 anti-CD34 monoclonal antibody¹⁸ (Becton Dickinson, San José CA).

RESULTS

Patients

One hundred and eighty nine patients were given an NST using the Mexican preparative regimen in the period of the study; of these, twelve individuals were given a 2-SCT; all these patients could not afford an allograft using conventional, myeloablative conditioning. The table 1 depicts the salient features of these 12 patients. Eight patients had a malignant condition (six acute leukemias, one myelofibrosis and one myelodysplasia); eleven patients were allografted twice from the same donor and in one case, two different cord blood cells were used. In two cases (numbers three and eight) the patient was initially allografted with umbilical cord blood cells from a sibling and later on with peripheral blood stem cells from the same sibling, already grown.

First allografts

The five patients with non malignant conditions were transfusion-dependent when the allograft was conducted; four out of six patients with leukemia were in a complete remission (CR) and two had refractory forms of the disease. Five patients failed to engraft and seven patients relapsed 2 to 13 months after the first allograft, see table 1. All the patients who failed to engraft did not develop chimerism; the seven patients who engrafted became full chimeras 30-60 days after the allograft

Second allografts

The median time to conduct the 2-SCT after the first one was 6 months (range 1-41). The five patients who failed to engraft after the first transplant failed also to engraft after the second one; all of

Table 1. Salient features of the twelve patients who received a second allogeneic stem cell transplantation.

Patient Number	Age/sex	Diagnosis, status	HSC source	Outcome, aGVHD	Remission (mos), cGVHD	Interval between transplants (mos)	HSC source	Outcome, aGVHD	Status, time (mos) cGVHD
		First allograft					Second allograft		
1	4/M	ALL, CR2	PB	CR, rel	13	16	PB	Ref	D, 2
2	21/M	ALL, ref	PB	CR, rel	2	4	PB	Ref, GI	D, 2
3	1/M	Thalassemia, TD	UCB	Failure	-	25	PB	Failure, TD	A, 37
4	49/F	AML-M2, CR1	PB	CR, rel	4	7	PB	CR, GI	A, 41, Ltd
5	9/M	ALL, CR2	PB	CR, rel	4	5	PB	Failure	D, 6
6	47/F	Myelofibrosis, TD	PB	Failure	-	1	PB	Failure	D, 1
7	9/M	ALL, ref	PB	CR, rel	3	3	PB	Failure	D, 1
8	3/M	ALL, CR2	UCB	CR, rel	13	22	PB	CR, rel	A, 10
9	8/F	Myelodysplasia, TD	PB	Failure	-	41	PB	Failure, TD	A, 35
10	24/F	AA, TD	PB	CR, rel, GI	8, Ltd	9	PB	CR, GI	A, 52
11	2/M	PRCA, TD	PB	Failure	-	12	PB	Failure, TD	A, 12
12	1/F	Osteopetrosis, TD	UCB	Failure	-	5	UCB	Failure, TD	A, 16

HSC: hematopoietic stem cells, aGVHD: acute graft *versus* host disease, mos: months, cGVHD: chronic graft *versus* host disease, M: male; f: female, CR: complete remission; CR1 or CR2: first or second complete remission, TD: transfusion dependent, ref: refractory, AA: aplastic anemia, PRCA: pure red cell aplasia; PB: peripheral blood; UCB: umbilical cord blood, rel: relapse, D: death, A: alive; GI: grade I acute GVHD; Ltd: limited chronic GVHD.

them but two (patient 6, with myelofibrosis and patient 9, with myelodysplasia), had benign conditions and had been heavily transfused before the allografts. Interestingly, four out of these individuals are alive, receiving transfusions. The 100-day mortality after the 2-SCT was 42% (5 of 12 patients); in three instances as a result of failure to engraft and in two cases, both patients with acute lymphoblastic leukemia, as the result of refractoriness of the malignancy. The two patients (numbers 2 and 7) allografted with refractory forms of lymphoblastic leukemia achieved very short remissions after the first allograft and failed to engraft after the second one, dying 2 and 1 months after the re-graft (Table 1). Only three patients (numbers 4, 8 and 10) were successfully “rescued” with the 2-SCT; in two instances patients with a relapsed acute leukemia (patients 4 and 8) and in the other a patient with a relapsed aplastic anemia (patient number 10).

Seven of the twelve patients are alive 10-41 months (median 35) after the 2-SCT; however, only three of them (25%) remain disease-free. The 52-month overall survival (SV) of the patients is 58%, whereas the median overall SV has not been reached, being above 52 months. The relapse-free survival of the patients is 25% at 52 months. All the patients who failed to engraft did not develop chimerism; patients 4, 8 and 10 are full chimeras. Figure 1 includes these data.

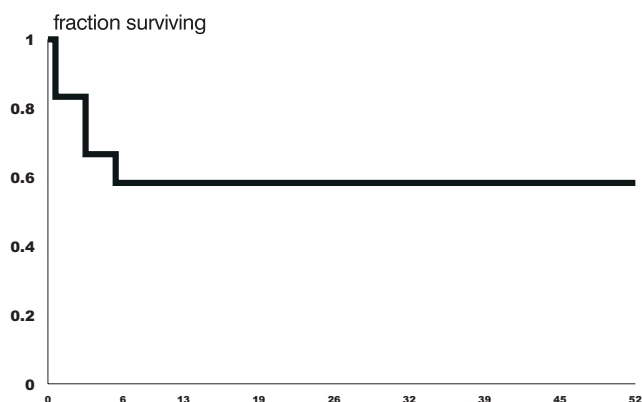


Figure 1. Overall survival after the second stem cell allograft of the 12 patients given two allotransplants using a non-myeloablative preparative regimen.

DISCUSSION

An allogeneic 2-SCT for hematological diseases recurring after first transplant may be a reasonable treatment option in selected patients; however, identification of factors influencing outcome and mortality after second transplantation and determination of patients groups who best respond to this therapy have proven difficult.² We have previously made comments on the salient features of the “Mexican” non-myeloablative preparative regimen to conduct

allogeneic stem cell transplantation.⁵⁻¹⁴ As most RIC regimens, the method relies mainly on the induction of a graft *versus* tumor effect, which is useful in some, but not all malignancies; in addition, the method has been shown to be useful to replace the hematopoietic tissue in circumstances in which this is needed.^{5,19-20} In this cohort of patients, only two with a malignancy (numbers 4 and 8) could be rescued by the 2-SCT. In case 4 by means of inducing a graft *versus* leukemia effect using the same donor, with a reduced and shortened immunosuppressive schedule; this patient had relapsed of the myeloid leukemia four months after the initial allograft and could be rescued with the 2-SCT using the same sibling donor,¹² remaining disease-free 41 months after the 2-SCT. The other patient was a boy with acute lymphoblastic leukemia in a second remission, who was initially grafted with umbilical cord blood cells from his sibling²¹ and, after another relapse, grafted with peripheral blood stem cells from the same sibling, 22 months old by then. Within patients with non-malignant conditions, patient number 10, with aplastic anemia, relapsed 8 months after the initial allograft and could be rescued with the 2-SCT of the same donor, by using a higher number of peripheral blood hematopoietic stem cells.¹⁹⁻²⁰

It has to be mentioned that the group of patients which we are hereby reporting is a highly selected one: included are individuals who lived enough after the first allograft in order to be re-allografted. By the same token, the prevalence of acute graft *versus* host disease (aGVHD) after the first allograft in this group of patients is very low (8%), this figure being substantially lower than that of the patients allografted using the "Mexican approach", in which the prevalence of aGVHD is around 30%.⁵⁻¹⁴ This may be related to both the failures to engraft after the initial transplant and also to the relapse rate in this group of patients. After the first allograft, chronic GVHD was observed in 1 out of 5 patients who engrafted and were followed by more than 100 days, whereas it was recorded in 1 out of 3 patients after the 2-SCT; these figures are also lower than that observed in the whole group of patients grafted using our method, which is around 50%.⁵⁻¹⁴ Along the same line, the 100-day mortality after the 2-SCT in this cohort was 42%, a figure significantly higher than that found in patients allografted using the "Mexican" conditioning regimen, which is around 10%.⁵⁻¹⁴ It is clear that the differences observed between this subset of patients and the whole group of individuals allografted using our method stem from the selection of the patients who can be given a 2-

SCT, selection which depends on features of both the disease and the patients, but which place these selected individuals in advantage as far as prognosis is concerned.

Despite the fact that the long-term disease-free SV in this group of patients given two consecutive stem cell allografts is rather poor (25%), the overall SV is acceptable (58% overall SV at 52 months), this figure being similar to that described by other authors conducting allogeneic 2-SCT.^{2,3} The overall SV figure could be related to the selection of patients as already discussed, but also to the low toxicity of the preparative regimen, which allows not only recovery of the endogenous hemopoiesis provided a graft failure occurs, but also repeated transplantation.⁵

In conclusion, conducting a 2-SCT in a given patient may be useful to rescue some individuals relapsing after a first hematopoietic allotransplant. Despite the fact that the success rate of this therapeutic maneuver is relatively low, it may be offered to selected patients. Conducting the allografts with a RIC conditioning allows re-allografting, which may benefit some patients.

REFERENCES

1. Thomas ED. Hematopoietic stem cell transplantation. *Sci Am* 1995; 272: 38.
2. Eapen M, Giralt SA, Horowitz MM, Klein JP, Wagner JE, Zhang MJ, et al. Litzow MR, deLima M. Second transplant for acute and leukemia relapsing after first HLA-identical sibling transplant. *Bone Marrow Transpl* 2004; 34: 721-7.
3. Guardiola P, Kuentz M, Garban F, Blaise D, Reiffers J, Attal M, et al. Second early allogeneic stem cell transplantations for graft failure in acute leukemia, chronic myeloid leukemia and aplastic anaemia. *Br J Haem* 2000; 111: 292-302.
4. Martino R, Caballero MD, De la Serna J, Díez-Martín JL, Urbano-Ispizua A, Tomás JF, et al. Low transplant-related mortality after second allogeneic peripheral blood stem cell transplant with reduced-intensity conditioning in adult patients who have failed a prior autologous transplant. *Bone Marrow Transpl* 2002; 30: 63-8.
5. Ruiz-Argüelles GJ, Gómez-Almaguer D. Braking dogmata to help patients: Non-myeloablative hematopoietic stem cell transplantation. *Expert Opin Biol Ther* 2004; 4: 1693-9.
6. Ruiz-Argüelles GJ, Ruiz-Argüelles A, Pérez-Romano B, Marín-López A, Delgado-Lamas JL. Non-cryopreserved peripheral blood stem cells autotransplants for hematological malignancies can be performed entirely on an outpatient basis. *Am J Hematol* 1998; 58: 161-4.
7. Ruiz-Argüelles GJ, López-Martínez B, Manzano C, Gómez-Rangel JD, Lobato-Mendizábal E. Significance of one human leukocyte antigen mismatch on outcome of non-myeloablative allogeneic stem cell transplantation from related donors using the Mexican schedule. *Bone Marrow Transpl* 2005; 35: 335-9.
8. Ruiz-Argüelles GJ, Gómez-Rangel JD, Ponce-de-León S, González-Déctor L, Reyes-Núñez V, Garcés-Eisele J. The Mexican schedule to conduct allogeneic stem cell transplantation is related to a low risk of cytomegalovirus reactivation and disease. *Am J Hematol* 2004; 75: 200-4.

9. Gómez-Almaguer D, Ruiz-Argüelles GJ, Ruiz-Argüelles A, González-Llano O, Cantú OE, Hernández NE. Hematopoietic stem cell allografts using a non-myeloablative conditioning regimen can be safely performed on an outpatient basis. *Bone Marrow Transpl* 2000; 25: 131-3.
10. Ruiz-Argüelles GJ, Gómez-Almaguer D, Ruiz-Argüelles A, González-Llano O, Cantú OG, Jaime Pérez JC. Results of an outpatient-based stem cell allotransplant program using non-myeloablative conditioning regimens. *Am J Hematol* 2001; 66: 241-4.
11. Ruiz-Argüelles GJ. Outpatient programs of myeloablative chemotherapy, autologous and allogeneic bone marrow transplantation. *Haematologica* 2000; 85: 1233-4.
12. Ruiz-Argüelles GJ, Gómez-Almaguer D, Gómez Rangel JD, Vela-Ojeda J, Cantú-Rodríguez OG, Jaime-Pérez JC, et al. Allogeneic hematopoietic stem cell transplantation with non-myeloablative conditioning in patients with acute leukemia eligible for conventional allografting: a prospective study. *Leukemia Lymphoma* 2004; 45: 1191-5.
13. Gómez-Almaguer D, Ruiz-Argüelles GJ, Tarín-Arzaga LC, González-Llano O, Jaime-Pérez JC, López-Martínez B, et al. Reduced-intensity stem cell transplantation in children and adolescents: The Mexican experience. *Biol Blood Marrow Transpl* 2003, 9: 157-61.
14. Ruiz-Argüelles GJ. Allogeneic stem cell transplantation using non-myeloablative conditioning regimens: Results of "the Mexican approach". *Int J Hematol* 2002; 76(Suppl. 1): 376-9.
15. Ruiz-Argüelles GJ, Gómez-Almaguer D, López-Martínez B, Cantú-Rodríguez OG, Jaime-Pérez JC, González-Llano O. Results of an allogeneic non-myeloablative stem cell transplantation program in patients with chronic myelogenous leukemia. *Haematologica* 2002; 87: 894-6.
16. Kessinger A, Armitage JO, Landmark JD, Smith DM, Weisenburger DD. Autologous peripheral hematopoietic stem cell transplantation restores hemopoietic function following marrow ablative therapy. *Blood* 1988; 71: 723-7.
17. Ruiz-Argüelles A. Flow cytometry in the clinical laboratory. Principles, applications and problems. *Ann Biol Clin* 1992; 50:735-43
18. Ruiz-Argüelles A, Orfao A. Caracterización y evaluación de células totipotenciales en sangre periférica y médula ósea. In: Ruiz-Argüelles GJ, San-Miguel JF (eds.). Actualización en leucemias. México City: Editorial Médica Panamericana; 1996, p. 79-82.
19. Jaime-Pérez JC, Ruiz-Argüelles GJ, Gómez-Almaguer D. Haematopoietic stem cell transplantation to treat aplastic anemia. *Expert Opin Biol Ther* 2005; 5: 617-26.
20. Gómez-Almaguer D, Vela-Ojeda J, Jaime-Pérez JC, Gutiérrez-Aguirre CH, Cantú-Rodríguez OG, Sobrevilla-Calvo P, et al. Allografting in patients with severe aplastic anemia using peripheral blood stem cells and a fludarabine-based conditioning regimen: The Mexican Experience. *Am J Hematol* 2006 (in the press).
21. Ruiz-Argüelles GJ, Gómez-Rangel D, Sánchez-Anzaldo J, Ruiz-Argüelles A, Porrás-Ramírez G, Luis-López A. Trasplante de células de cordón umbilical: informe de dos casos. *Medicina Univ* 2002; 4: 233-5.

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