Published in final edited form as:
Bone Marrow Transplant. 2009 March ; 43(6): 477–480. doi:10.1038/bmt.2008.353.

Salvage transplantation for allograft failure using fludarabine and alemtuzumab as conditioning regimen

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Abstract

Graft failure after allogeneic blood or marrow transplantation, although generally uncommon, can be a devastating complication. This report includes the outcome of nine patients who received a salvage transplant for failure to engraft after one (n = 8) or 2 (n = 1) prior transplants. Eight patients received allografts from the original donor. All received fludarabine 30 mg/m^2 i.v. and alemtuzumab 20 mg i.v. daily from days −6 to −2. Daily CYA was begun on day −2, and the allograft was infused on day 0. The therapy was well tolerated with low toxicity, and all nine patients engrafted, recovering neutrophils at a median of 12 days after transplant. Four patients died: two of relapse, one of a fungal infection in the setting of GVHD and one of multiple sclerosis. The combination of fludarabine and alemtuzumab is an effective and well-tolerated salvage conditioning regimen for patients who experience graft failure after blood or marrow transplants.

Keywords

graft failure; alemtuzumab; fludarabine

Introduction

Graft failure is rare in recipients of allogeneic blood or marrow transplantation (BMT) from HLA-identical donors in the absence of T-cell depletion, but can occur in greater than 10% of T-cell-depleted or HLA mismatched transplants.[1,2] Graft failure can occur early before any evidence of neutrophil recovery or late after transient donor engraftment. In addition to T-cell depletion and HLA-mismatch, factors such as poor graft quality, reduced-intensity conditioning regimens and alloimmunization can also be associated with increased rates of graft failure, and in many cases the cause is probably multifactorial.[1–5] Regardless of the cause, the outcome with a second transplant has historically been rather poor because of high rates of infections, extramedullary organ toxicity, and GVHD.[2,6–16] Thus, we hypothesized that the optimal second BMT strategy for salvaging graft failure should involve substantial immunosuppression and high doses of donor cells, whereas at the same time limiting cytotoxicity and GVHD.

We report on a salvage transplant strategy using a conditioning regimen of alemtuzumab and fludarabine. These drugs are frequently administered in BMT conditioning regimens,

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especially those of reduced intensity, because of their potent immunosuppressive properties but limited cytotoxic properties. Alemtuzumab is an MoAb directed against CD52, and fludarabine, through its metabolites, acts as an inhibitory substrate for ribonucleotide reductase, the DNA polymerases, DNA ligase I and DNA primase secondarily affecting transcription through incorporation into RNA.[17,18] In addition, pretransplant alemtuzumab has an elimination half-life of approximately 12 days, and the antibody will still be present at the time of allograft infusion, thus generating some degree of *in vivo* T-cell depletion as GVHD prophylaxis.[18]

Materials and methods

This is a retrospective review of all patients who received a salvage allogeneic BMT for graft failure since September 2001 at the Sidney Kimmel Comprehensive Cancer Center at Johns Hopkins. The diagnosis of graft failure was made when patients beyond day 30 after allogeneic BMT failed to show both neutrophil recovery and any evidence of donor origin DNA in chimaerism studies by PCR of variable nucleotide tandem repeats. Regardless of the primary diagnosis, patients received fludarabine 30 mg/m^2 i.v. and alemtuzumab 20 mg i.v. daily from days −6 to −2. On day −2, they began CYA 5 mg/kg i.v. over 6 h every day. The allograft was infused on day 0. On day +3 after BMT, CYA was decreased to 2.5 mg/kg i.v. and then given orally when the patient could tolerate it for 6 months. On day +5 after BMT, 5 μg/kg G-CSF was given until neutrophil recovery to more than 500/μl. The standard antibiotic prophylaxis included sulphamethoxazole/trimethoprim (160 mg trimethoprim component or single strength) p.o. every day for 6 months starting on day 21, valacyclovir 500 mg. p.o. 3 times a day until day 28, fluconazole 400 mg p.o. every day and norfloxacin 400 mg p.o. twice a day until neutrophil recovery.

Results

Between September 2001 and March 2007, nine consecutive patients who failed to engraft after an allograft were re-transplanted (Table 1). The median time between failed transplant and salvage allograft was 62 days (range 43–84). All patients had a BM biopsy before the salvage transplant, and in all cases the marrow was aplastic except in one (patient number 2) who was hypocellular (<5%) and only showed persistent CLL but no haematopoietic progenitors. In eight patients, the salvage transplant followed their first allograft, whereas in another patient it followed after a second transplant. One patient (number 9) received tacrolimus (1 mg i.v./day starting on day −1) instead of CYA. All patients engrafted; eight exhibited 100% donor chimaerism by molecular studies around day 60, whereas one died with neutrophil recovery but before donor chimaerism studies were performed. Engraftment was rapid with neutrophils reaching 500/μl at a median of 12 days after transplantation (range 10–25). Three patients never recovered their platelets (platelet recovery was defined as the first day of a platelet count > 20 000/μl without a platelet transfusion in the preceding 7 days). Of the six who did, the median time to recovery was 21 days (range 16–46). One patient received a BM allograft and eight were transplanted with mobilized peripheral blood stem cells. Characteristics of the grafts are listed in Table 2. The outcome of the nine patients is listed in Table 3. There were two cases of GVHD: one patient had skin-only acute grade II, and the second had grade II acute and later chronic of the skin-mild chronic, according to the NIH consensus definition,19 and limited according to the Seattle definition.[20] Both patients responded to steroid-based therapy for the acute phase, and no therapy was given to the patient with chronic GVHD as she was asymptomatic and by now free of clinical manifestations. Of the nine patients, five are alive and in remission with a performance status of ECOG 0 at their last visit. One patient died of a fungal infection in the setting of GVHD therapy 37 days after BMT, one died of progressive multiple sclerosis without evidence of disease, and two died of relapse. No CMV reactivation was seen on any patient (seven as screened by polymerase chain

reaction in blood and two by pp65 antigen monitoring). Only one long-term survivor developed a probable fungal infection (pneumonia) that responded well to liposomal amphotericin. As of 30 June 2008, the median overall survival is 453 days (range 37–821).

Discussion

Historically, the outcome of patients with graft failure following myeloablative allogeneic BMT is poor.[2,6–9] In particular, the lengthy periods of neutropenia lead to high rates of severe infectious complications.[6] Rates of severe extramedullary end-organ toxicities and of GVHD are also high. The high extramedullary conditioning regimen toxicity rates probably are the result of using two conditioning regimens in a short period of time, whereas the high rate of GVHD is most likely multifactorial and includes intensive conditioning and attempts to enhance engraftment at the expense of GVHD.[18]

Multiple regimens have been used for second transplants for graft failure, but most have included high-dose CY because of its potent immunosuppressive properties.[6,9,13–15] However, its cytotoxic properties may be at least partly responsible for the high rate of conditioning regimen-related toxicity. Fludarabine and alemtuzumab are potent immunosuppressives with limited non-haematologic cytotoxic properties, which have led to their wide and effective use in reduced-intensity conditioning regimens. Moreover, as already discussed, pretransplant alemtuzumab has GVHD-modifying effects.[18] Recently, Chewning *et al.*[21] reported that 15 of 15 patients engrafted after undergoing a second BMT for graft failure with fludarabine-based conditioning regimens. Byrne *et al.*[22] also reported on 9 patients engrafting out of 11 patients treated with fludarabine, CY and alemtuzumab for a salvage BMT. Jabbour *et al.*[16] also reported on nine patients receiving a salvage BMT with fludarabine and antithymocyte globulin, six engrafting and developing GVHD, and there was only one survivor with extensive chronic GVHD. Ayas *et al.*[23] recently reported on four patients with Fanconi's anaemia receiving a salvage BMT after antithymocyte globulin; three engrafted and are long-term survivors, whereas encouraging the Ayas series is exclusively in Fanconi's anaemia patients and is thus not necessarily applicable to standard haematologic malignancy patients.

Fludarabine and alemtuzumab proved to be effective and well tolerated, with 100% donor engraftment in all patients, only one transplant-related death, and five of the nine patients surviving at a median of 453 days. Although larger trials, possibly comparing different salvage-conditioning regimens, would be ideal, the rarity of graft failure makes this unfeasible. Nevertheless, although rare, graft failure remains an important complication of allogeneic transplantation. Using this combination as salvage conditioning offers the potential advantage of excluding additional cytotoxic therapy and its attendant toxicities. The small patient numbers and heterogeneity of diseases preclude firm conclusions about the long-term disease control with this approach. However, the combination of myeloablation (from the failed BMT) and complete donor chimaerism suggests that these patients should fare no differently from similar patients engrafting after the initial myeloablative regimen.

Acknowledgments

This research was supported in part by the Grant CA15396 from the NIH. This study was presented in part at the 2008 Meeting of the American Society for Blood and Marrow Transplantation in San Diego, CA, USA.

References

1. Anasetti C, Amos D, Beatty PG, Appelbaum FR, Bensinger W, Buckner CD, et al. Effect of HLA compatibility on engraftment of bone marrow transplants in patients with leukemia or lymphoma. N Engl J Med 1989;320:197–204. [PubMed: 2643045]

2. Champlin RE, Horowitz MM, van Bekkum DW, Camitta BM, Elfenbein GE, Gale RP, et al. Graft failure following bone marrow transplantation for severe aplastic anemia: risk factors and treatment results. Blood 1989;73:606–613. [PubMed: 2644980]

3. Wagner JL, Deeg HJ, Seidel K, Anasetti C, Doney K, Sanders J, et al. Bone marrow transplantation for severe aplastic anemia from genotypically HLA-nonidentical relatives. An update of the Seattle experience. Transplantation 1996;61:54–61. [PubMed: 8560574]

4. Hansen JA, Gooley TA, Martin PJ, Appelbaum F, Chauncey TR, Clift RA, et al. Bone marrow transplants from unrelated donors for patients with chronic myeloid leukemia. N Engl J Med 1998;338:962–968. [PubMed: 9521984]

5. Storb R, Etzioni R, Anasetti C, Appelbaum FR, Buckner CD, Bensinger W, et al. Cyclophosphamide combined with anti-thymocyte globulin in preparation for allogeneic marrow transplants in patients with aplastic anemia. Blood 1994;84:941–949. [PubMed: 8043876]

6. Storb R, Weiden PL, Sullivan KM, Appelbaum FR, Beatty P, Buckner CD, et al. Second marrow transplants in patients with aplastic anemia rejecting the first graft: use of a conditioning regimen including cyclophosphamide and antithymocyte globulin. Blood 1987;70:116–121. [PubMed: 3297199]

7. Redei I, Waller EK, Holland HK, Devine SM, Wingard JR. Successful engraftment after primary graft failure in aplastic anemia using G-CSF mobilized peripheral stem cell transfusions. Bone Marrow Transplant 1997;19:175–177. [PubMed: 9116616]

8. Advisory Committee of the International Bone Marrow Transplant Registry. Bone marrow transplantation from donors with aplastic anemia. A report from the ACS/NIH bone marrow transplant registry. JAMA 1976;236:1131–1135. [PubMed: 781333]

9. Stucki A, Leisenring W, Sandmaier BM, Sanders J, Anasetti C, Storb R. Decreased rejection and improved survival of first and second marrow transplants for severe aplastic anemia (a 26-year retrospective analysis). Blood 1998;92:2742–2749. [PubMed: 9763558]

10. Kersey J, Roloff J, Hokeness S, Johnson FL, Nesbit M, Krivit W. Retransplantation of human bone marrow. Two immuno-suppressive drug regimens. Transplantation 1975;19:475–478. [PubMed: 237352]

11. Meuwissen HJ, Moore EC, Strauss HS, Taft E, Britten A. Successful retransplantation of bone marrow following failure of initial engraftment in a patient with aplastic anemia. J Pediatr 1976;89:588–592. [PubMed: 784930]

12. Gmur J, Frick PG, von Felten FA. Successful allogeneic bone marrow retransplantation with the same donor after graft rejection: application of a modified conditioning regimen. Blut 1979;39:47–52. [PubMed: 380689]

13. Al-Qurashi F, Ayas M, Al Sharif F, Ibrahim E, Sahovic E, Al Mahr M, et al. Second allogeneic bone marrow transplantation after myeloablative conditioning analysis of 43 cases from single institution. Hematology 2004;9:123–129. [PubMed: 15203867]

14. Grandage VL, Cornish JM, Pamphilon DH, Potter MN, Steward CG, Oakhill A, et al. Second allogeneic bone marrow transplants from unrelated donors for graft failure following initial unrelated donor bone marrow transplantation. Bone Marrow Transplant 1998;21:687–690. [PubMed: 9578308]

15. Radich JP, Sanders JE, Buckner CD, Martin PJ, Petersen FB, Bensinger W, et al. Second allogeneic marrow transplantation for patients with recurrent leukemia after initial transplant with total-body irradiation-containing regimens. J Clin Oncol 1993;11:304–313. [PubMed: 8426208]

16. Jabbour E, Rondon G, Anderlini P, Giralt SA, Couriel DR, Champlin RE, et al. Treatment of donor graft failure with nonmyeloablative conditioning of fludarabine, antithymocyte globulin and a second allogeneic hematopoietic transplantation. Bone Marrow Transplant 2007;40:431–435. [PubMed: 17603511]

17. Elter T, Hallek M, Engert A. Fludarabine in chronic lymphocytic leukaemia. Expert Opin Pharmacother 2006;7:1641–1651. [PubMed: 16872267]

18. Bolaños-Meade J, Vogelsang GB. Acute graft-versus-host disease. Clin Adv Hematol Oncol 2004;2:672–682. [PubMed: 16163254]

19. Filipovich AH, Weisdorf D, Pavletic S, Socie G, Wingard JR, Lee SJ, et al. National Institutes of Health consensus development project on criteria for clinical trials in chronic graft-versus-host

disease: I. Diagnosis and staging working group report. Biol Blood Marrow Transplant 2005;11:945–956. [PubMed: 16338616]

20. Shulman HM, Sullivan KM, Weiden PL, McDonald GB, Striker GE, Sale GE, et al. Chronic graft-versus-host syndrome in man. A long-term clinicopathologic study of 20 Seattle patients. Am J Med 1980;69:204–217. [PubMed: 6996481]
21. Chewning JH, Castro-Malaspina H, Jakubowski A, Kernan NA, Papadopoulos EB, Small TN, et al. Fludarabine-based conditioning secures engraftment of second hematopoietic stem cell allografts (HSCT) in the treatment of initial graft failure. Biol Blood Marrow Transplant 2007;13:1313–1323. [PubMed: 17950918]
22. Byrne BJ, Horwitz M, Long GD, Gasparetto C, Sullivan KM, Chute J, et al. Outcomes of a second non-myeloablative allogeneic stem cell transplantation following graft rejection. Bone Marrow Transplant 2008;41:39–43. [PubMed: 17982503]
23. Ayas M, Al-Jefri A, Al-Seraihi A, Al-Mahr M, Al-Ahmari A, El-Solh H. Second stem cell transplantation in patients with Fanconi anemia using antithymocyte globulin alone for conditioning. Biol Blood Marrow Transplant 2008;14:445–448. [PubMed: 18342787]
24. Santos GW, Tutschka PJ, Brookmeyer R, Saral R, Beschorner WE, Bias WB, et al. Marrow transplantation for acute nonlymphocytic leukemia after treatment with busulfan and cyclophosphamide. N Engl J Med 1983;309:1347–1353. [PubMed: 6355849]
25. Wingard JR, Piantadosi S, Santos GW, Saral R, Vriesendorp HM, Yeager AM, et al. Allogeneic bone marrow transplantation for patients with high-risk acute lymphoblastic leukemia. J Clin Oncol 1990;8:820–830. [PubMed: 2332770]
26. Luznik L, Fuchs EJ, Chen AR, Kaup M, Bright EC, Bolaños-Meade J, et al. Post-transplantation high-dose cyclophosphamide (Cy) is effective single agent GVHD prophylaxis that permits prompt immune reconstitution after myeloablative HLA matched related and unrelated bone marrow transplantation (BMT). Biol Blood Marrow Transplant 2007;13:4.
27. Luznik L, O'Donnell PV, Symons HJ, Chen AR, Leffell MS, Zahurak M, et al. HLA-haploidentical bone marrow transplantation for hematologic malignancies using nonmyeloablative conditioning and high-dose, posttransplantation cyclophosphamide. Biol Blood Marrow Transplant 2008;14:641–650. [PubMed: 18489989]

Table 1

Demographic information

Patient	Gender	Age at BMT	Diagnosis	Disease status at BMT	Time from BMT to salvage BMT
1	M	41	CMML	Refractory	81 days
2	M	46	CLL	PR	60 days
3	F	52	HL	CR	48 days
4	F	47	AML	CR2	84 days
5	M	31	NHL	PR	62 days
6	F	50	MDS	Untreated	43 days
7	F	50	CML	CP	69 days
8	F	60	CMML	Untreated	81 days
9	F	63	AML	CR	62 days

Abbreviations: CMML = chronic myelomonocytic leukaemia; CP = chronic phase; CR = complete response; F = female; HL = Hodgkin's lymphoma; M = male; MDS = myelodysplastic syndrome; NHL = non-Hodgkin's lymphoma; PR = partial response.

Table 2

Information of the grafts

Patient	Salvage alloBMT	Prior BMT	Same donor as previous BMT	Salvage graft	CD34$^+$/kg	CD3$^+$	Patient/donor ABO
1	Second transplant	BU/CY[a] MUD BMT	Yes	MUD BM	4.90E + 06[b]	NA	A + /O +
2	Second transplant	CY/TBI HLA identical sibling BMT[c]	Yes	Allo PBSC	5.63E + 06	2.10E + 08	A + /A +
3	Second transplant	BU/CY HLA identical sibling BMT[a]	Yes	Allo PBSC	5.46E + 06	4.10E + 08	O + /B +
4	Second transplant	BU/CY HLA identical sibling BMT[d]	Yes	Allo PBSC	2.61E + 07	9.93E + 08	A + /A−
5	Second transplant	BU/CY HLA identical sibling BMT[e]	Yes	Allo PBSC	2.31E + 06	2.33E + 08	A + /O +
6	Second transplant	BU/CY HLA identical sibling BMT[d]	No	Allo PBSC	2.56E + 06	4.45E + 08	B + /B +
7	Third transplant	Flu/CY/TBI haploBMT[f]; then BU/CY[d] MUD BMT	Yes (different from 1st BMT)	MUD PBSC	9.35E + 06	4.69E + 08	O + /O +
8	Second transplant	BU/CY[d] MUD BMT	Yes	MUD PBSC	7.58E + 06	5.65E + 08	B + /O +
9	Second transplant	BU/CY[d] MUD BMT	Yes	MUD PBSC	3.60E + 08	2.37E + 07	O−/O +

Abbreviations: Allo = allogeneic; Flu = fludarabine; haplo = haploidentical; MUD = matched unrelated donor.

[a] BU (16 mg/kg) plus CY (200 mg/kg) as described by Santos *et al.*[24]

[b] Nucleated cells/kg.

[c] CY (200 mg/kg) plus TBI 1200 cGy as described by Wingard *et al.*[25]

[d] BU (16 mg/kg), CY (100 mg/kg) plus post transplant CY (100 mg/kg) as described by Luznik *et al.*[26]

[e] BU (16 mg/kg) plus CY (200 mg/kg) with T-cell-depleted graft.

[f] Fludarabine (120 mg/m^2), CY (29 mg/kg), TBI 200 cGy, plus post transplant CY (100 mg/kg) as described by Luznik *et al.*[27]

Table 3

Outcome of the patients

Patient	Chimaerism day 60	Chimaerism day 180	Status	Cause of death	GVHD	Days to neutrophil recovery
1	100% donor BM	NA	Dead	Relapse	No	25
2	100% donor PB	NA	Dead	Relapse	No	12
3	NA (neutrophil recovered)	NA	Dead	Fungal infection	Yes (skin)	14
4	100% donor PB	100% donor PB	Dead	Primary progressive multiple sclerosis	No	10
5	100% donor PB	100% donor BM	CR	Alive	No	10
6	100% donor PB	100% donor BM	CR	Alive	No	11
7	100% donor PB	100% donor PB	CR[a]	Alive	No	12
8	100% donor PB	100% donor BM	CR	Alive	Yes (skin)	10
9	100% donor PB	100% donor BM	CR	Alive	No	12

Abbreviations: aGVHD = acute GVHD; NA = not available (chimaerism determination was not available, but the patient had neutrophil recovery).

[a]Thirteen months after her salvage transplant, she became a mixed chimaera with 10% Ph1 positive cells by FISH, and was given dasatinib, thus becoming a 100% donor again 3 months later.