Non-myeloablative allogeneic hematopoietic cell transplantation for relapsed or refractory Waldenström macroglobulinemia: evidence for a graft-*versus*-lymphoma effect

Irrespective of age, patients with aggressive Waldenström macroglobulinemia (WM) who have exhausted chemo-immunotherapy-based approaches may be candidates for allogeneic hematopoietic cell transplantation (HCT), a strategy that has been rarely attempted.[1-7] In Seattle, conditioning with a single fraction of low-dose total body irradiation (TBI), combined with a graft-*versus*-host disease (GvHD) prophylaxis consisting of cyclosporine and mycophenolate mofetil, led to near-uniform allogeneic engraftment in a canine DLA-identical model.[8] Clinical trials based on this approach demonstrated rapid engraftment and graft-*versus*-tumor effects in a wide variety of hematologic malignancies, with higher sustained response rates for patients with indolent diseases.[9] Here we present a retrospective analysis of outcomes among 15 heavily pre-treated and largely chemo-refractory WM patients who received a minimal intensity conditioning regimen consisting of 200 cGy TBI ± fludarabine, in preparation for HLA-matched

Table 1. Outcomes of hematopoietic cell transplantation.

UPN	Donor	Age at HCT	Time Dx -HCT (years)	Lines, n	ChR	HCT-CI	Allogeneic HCT conditioning regimen	Grade acute GvHD	Extensive chronic GvHD	Best response after HCT (years)	Survival (years)	Current status
1	MRD	50	2.9	3	+	2	TBI	II	+	IFIX-neg CR (2.6)	6.3	Died from relapsed WM in nodes.
2	MRD	58	10.3	2	+	0	TBI	II	_	VGPR (6.7)	16.2	Died from relapsed WM 12.5 years after HCT.
3	MUD	53	15.4	9	+	3	Flu/TBI	II	_	PR	0.8	Died from host-derived AML
4	MRD	57	1.8	2	_	2	BEAM Auto / TBI	0	+	CR (1.0)	1.3	Died from relapsed DLBCL. IFIX not done.
5	MRD	61	7.0	4	+	2	Flu/TBI	0	_	IFIX-neg CR (0.5)	15.8 +	Alive in CR. Off IST. KS: 100%.
6	MRD	54	1.8	2	+	1	Flu/TBI	III	+	PR	4.8	Died from infection on IST for chronic GvHD.
7	MUD	44	1.5	2	+	0	Flu/TBI	I	NE	NE	0.1	Died at day 29 from aspiration pneumonia
8	MUD	63	6.6	6	+	0	Flu/TBI	II	+	IFIX-neg CR (0.9)	12.1 +	Alive in CR. Off IST. KS: 100%.
9	MRD	57	9.5	5	_	0	CY-TBI Auto / TBI	II	+	IFIX-neg CR (2.1)	11.9 +	Alive in CR. Off IST. KS: 90%.
10	MUD	64	19.2	5	_	3	Flu/TBI	II	+	IFIX-neg CR (6.9)	11.3 +	Alive in CR. Off IST. KS: 90%.
11	MUD	53	4.4	4	_	3	Flu/TBI	0	+	IFIX-neg CR (0.8)	12.1 +	Alive in CR. Off IST. KS: 90%.
12	MRD	65	7.2	5	+	2	Flu/TBI	0	NE	PR	0.4	Rejected his graft and died with pancytopenia.
13	MRD	53	4.4	7	+	0	Flu/TBI	0	_	IFIX-neg CR (0.7)	9.6 +	Alive in CR. Off IST. KS: 90%.
14	MUD	68	8.0	5	+	3	Flu/TBI	I	+	IFIX-neg CR (1.6)	2.2	Died from metastatic melanoma. Marrow not done.
15	MRD	51	1.3	3	_	3	BEAM Auto / TBI	II	_	PR	6.6 +	Alive in PR, with improving IgM and M-spike. Off IST. KS: 90%

UPN: unique patient number; HCT: hematopoietic cell transplantation; Dx: diagnosis; n: number; ChR: chemo-refractoriness; HCT-CI: hematopoietic cell transplantation comorbidity index; GvHD: graft-*versus*-host disease; MRD: matched related donor; MUD: matched unrelated donor; TBI: total body irradiation; Flu: fludarabine; BEAM: carmustine, cytarabine, etoposide, melphalan; Auto: autologous hematopoietic cell transplantation; CY: cyclophosphamide; NE: not evaluable; IFIX: immunofixation; CR: complete remission; VGPR: very good partial remission; PR: partial remission; WM: Waldenström macroglobulinemia; AML: acute myeloid leukemia; DLBCL: diffuse large B-cell lymphoma; IST: immunosuppressive therapy; KS: Karnofsky performance status scale.

related or unrelated HCT.

Between March 1999 and June 2010, 15 patients with WM received allogeneic HCT at the Fred Hutchinson Cancer Research Center (Seattle, WA, USA), Oregon Health & Science University, (Portland, OR, USA) and the University of Leipzig (Germany), on prospective trials coordinated through the Fred Hutchinson Center and approved by each organization's institutional review board. The trials were registered at "*clinicaltrials.gov*". The 15 patients had received a median of four (range, 2-9) prior lines of chemotherapy (Table 1). At the time of transplantation, 10 of the 15 patients had chemo-refractory disease. None of the patients had received a Bruton kinase inhibitor. Two patients had been given proteasome inhibitors, one of whom had a partial response. Four patients received multiple plasmaphereses before HCT, and 2 of the 4 continued on plasmaphereses for six months after HCT. Three patients received planned cytoreductive autologous HCT before allogeneic HCT, either to treat co-existing transformed diffuse large B-cell lymphoma (n= 1) or to reduce large tumor burden (n=2). Donors were HLA-identical siblings in 9 cases and HLA-matched (at the allele-level) unrelated in 6 cases. All patients received conditioning with 200 cGy TBI at 7 cGy/min from a linear accelerator or opposing cobalt-60 source on day 0. In addition, 13 patients were given three doses of fludarabine, 30 mg/m^2/day, on days -4 to -2. One patient received four doses of rituximab (first dose on day -3 and the last three after HCT). Granulocyte colony-stimulating factor-mobilized peripheral blood stem cells (3.96 to 16.48 × 10^6 CD34$^+$ cells/kg) were infused on day 0 after TBI in 14 patients, while one patient received a marrow graft. Post-grafting immunosuppression included mycophenolate mofetil (28 days for patients with sibling donors and 100 days for recipients of grafts from unrelated donors) and a calcineurin inhibitor, either cyclosporine or tacrolimus, for 180 days; one patient was also given sirolimus for GvHD prophylaxis, as per protocol. Thirteen patients had prompt and sustained engraftment with normalization of blood counts. One patient died on day 29 from respiratory insufficiency and was not evaluable for sustained engraftment. One patient rejected his graft and died with pancytopenia from bacterial infection on day 150; this patient had been on prednisone, 20-50 mg/day, before and after HCT for treatment of presumed fludarabine-associated autoimmune hemolytic anemia. The cumulative incidence of grade II-III acute GvHD was 70% at a median of 48 days (range, 15-119). No grade IV acute GvHD was observed. In all patients acute GvHD resolved after one course of methylprednisolone. The cumulative incidence of extensive chronic GvHD was 62% at a median of 5.5 months (range, 3-50). In all but 2 of the surviving patients, therapy for chronic GvHD has been successfully discontinued. Five-year mortality from causes other than WM was 40%. One patient with refractory WM and poor performance status died on day 29 after HCT due to generalized seizures and respiratory insufficiency. One patient with extensive chronic GvHD died of pneumonia. The patient who rejected the graft died from infection. One patient in remission from WM died of relapse of pre-existing transformed DLBCL.

Two cases of non-relapse mortality resulted from secondary malignancies (Table 1): one from host-derived acute myeloid leukemia and one from metastatic malignant melanoma diagnosed 1 year after HCT; the latter patient had concurrent biopsy-proven liver involvement with host-derived, Epstein-Barr virus-negative, anaplastic large cell CD30$^+$ ALK-negative lymphoma whose origin (primary *versus* WM-transformed) could not be ascertained. All evaluable patients showed disease responses (Table 2).

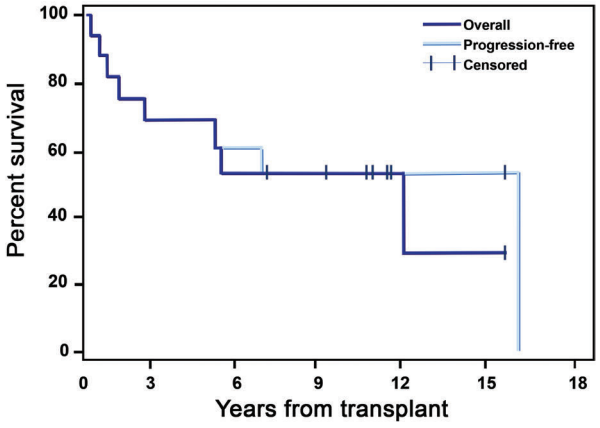

Figure 1. Kaplan-Meier curves for overall and progression-free survival.

Eight achieved immunofixation-negative complete remissions (57%), one had an immunofixation-unconfirmed complete remission, one achieved a very good partial remission, and 4 experienced partial remission. The median time to best response was 1.3 years (range, 0.5-7). Three patients achieved complete remissions 2.4, 3.2, and 14.7 months after completing the scheduled GvHD prophylaxis (n=1) or immunosuppressive treatment for chronic GvHD (n=2), as shown by gradual normalization of serum IgM levels and disappearance of the M-spike throughout the post-transplant course. At last contact, 7 patients are alive, 6 in complete remission and one in improving partial remission. With a median follow up of 11.3 years (range, 6.6-15.8) among surviving patients, the estimated 10-year overall and progression-free survival rates were both 53% (95% CI: 0.26-0.74) (Figure 1).

The 15 patients with advanced WM in the current exploratory study were treated with allogeneic HCT at a median of 7.2 years after diagnosis and following a median of four lines of chemotherapy. Ten of the 15 were considered chemotherapy-refractory. Given that some of the patients were older while others were frail, in part owing to many years of chemotherapy, we chose a minimal-intensity conditioning regimen for HCT. While the associated toxicity was mild and all patients remained in the outpatients care setting through the early post-transplant period, the intensity of the regimen did not lend itself to achieving significant tumor cell killing. Therefore, success with this transplant approach almost entirely reflected an allogeneic graft-*versus*-tumor effect exerted by donor T cells. One limitation of this approach is the requirement of a minimum of 6 months of post-grafting immunosuppressive therapy to control GvHD, which also attenuates graft-*versus*-tumor effects. Despite this immunological "handicap," 8 of the 13 evaluable patients achieved complete remission, which has been maintained in 6 for 9.6 to 15.8 years up to now. These remissions did not follow a predictable pattern, with deepest responses occurring over a broad range of time between 6 months and almost 7 years after transplantation. Of note, 6 of the 8 patients who achieved complete remission experienced chronic GvHD that resolved after immunosuppressive therapy in 5 of them. Three of the patients who are alive in complete remission had chemotherapy-sensitive WM and 3 had chemotherapy-resistant disease. The seventh complete remission patient (patient #1) relapsed 5.1 years after HCT while on immunosuppressive therapy for chronic GvHD and died at 6.3 years. The eighth patient who achieved complete remission remained in remission from WM but relapsed with a pre-existing, concurrent dif-

Table 2. Disease burden at allogeneic transplantation and best results after transplantation.

UPN	IgM pre-HCT (mg/dL)	IgM best	M-spike pre-HCT (g/dL)	M-spike best pre-HCT	BM pre-HCT (%)	BM best	Adenopathy pre-HCT	Adenopathy post-HCT
1	697	44	0.5	0	30	0	–	–
2	13000[¥]	400	2.1	0.4	20	0	–	–
3	11900[¥]	1810	5.6	3.1	65	7	–	–
4	570	87	0.1	0	10 *	0	Positive	Persistent
5							–	–
6	3400	236	1.5	0.2	25	3	–	–
7	3960	NE	2.0	NE	75	NE	Positive	NE
8	1370	265	1.0	0	45	0	Bulky	Resolved
9	845	53	0.5	0	5	0	Bulky	Resolved
10	2470	52	1.4	0	25	0	Positive	Resolved
11	897	26	TSTQ	0	1	0	Positive	Resolved
12	1222[¥]	219	0.6	0.2	10	0	Bulky	Persistent
13	4780[¥]	197	3.2	0	7	0	–	–
14	2810	100	1.5	0	70	40	–	–
15	1550	617	1.5	0.4	20	1	–	–

UPN: unique patient number; HCT: hematopoietic cell transplantation; BM: marrow lymphoplasmacytic infiltration; TSTQ: too small to quantitate; NE: not evaluable; Bulky: any lymphadenopathy with a transverse diameter of more than 5 cm as measured by computed tomography; Positive: any lymphadenopathy with a transverse diameter of less than 5 cm as measured by computed tomography. *Bone marrow histopathological examination was negative for DLBCL cells. [¥]Patients who required plasmapheresis to control hyperviscosity syndrome before allogeneic transplantation.

fuse large B-cell lymphoma 1.3 years after HCT, with the latter cells outgrowing the anti-tumor effect of the donor T cells.

Owing to the attenuated immune responses early after HCT resulting from both the very gradual conversion to a donor-derived immune system and the broad immune suppression from drugs aimed at GvHD prevention, most patients experienced an extended period of mixed donor-host hematopoietic chimerism.[10] While mixed chimerism may not have adverse consequences for patients with low tumor burden before HCT, it may be permissive for relapse among patients whose tumors grow quickly. Examples in the current study include patient #4 with diffuse large B-cell lymphoma, patient #3 who likely had therapy-induced acute myeloid leukemia, and patient #14 with CD30$^+$ anaplastic large cell lymphoma, even though the proximate cause of death was metastatic melanoma diagnosed 1 year after HCT. The coexistence of second cancers in patients with WM is not surprising. A publication by Varettoni *et al.* estimated the increased risk of second cancers to be 1.69.[11]

Only one of the 15 patients (#12) failed to show sustained engraftment. This patient had been receiving prednisone for autoimmune hemolytic anemia. Graft failure is likely attributable to the use of prednisone when viewed in the context of previous preclinical studies that had shown uniform graft failure in dogs given prednisone peri-transplantation,[12] although there exists a minimal risk of graft failure with minimal intensity conditioning.[13] Given that WM is otherwise incurable, it could be argued that allogeneic transplantation should be considered earlier in the course of this disease. That way, patients would be transplanted while their disease is still chemotherapy-sensitive, which promises to significantly lessen the risk of post-transplant relapse. Earlier transplantation might also reduce the risk of developing second cancers with aggressive morphology, such as was seen in 3 of our patients. Finally, patients would likely be in better general condition without having acquired cumulative toxicities from many years of chemotherapy.[14] This would significantly lessen the risk of post-transplant non-relapse mortality compared to that of patients with organ damage from extensive chemotherapy.

In conclusion, despite improvements in chemotherapeutic drugs, WM has remained an incurable illness. However, we and others have shown that allogeneic HCT can achieve cures in nearly half of the patients with advanced WM including those with chemotherapy-resistant disease. The risk of non-relapse mortality must be carefully evaluated before the procedure. It is possible that results of HCT can be further improved by transplanting earlier in the disease course when patients are in a better general condition or before they become refractory to chemotherapy or develop second cancers.

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