Tandem autologous-allogeneic stem cell transplantation as a feasible and effective procedure in high-risk lymphoma patients

Although high-dose chemotherapy is the gold standard for the treatment of many relapsed or refractory lymphomas, the outcome remains unsatisfactory, particularly in some subsets of patients with adverse prognostic features. Here we report the outcome of 111 high-risk lymphoma patients treated with a tandem strategy involving debulking with HDC followed by autologous stem cell transplantation (ASCT) and subsequent adoptive immunotherapy consisting of allogeneic stem cell transplantation (SCT; tandem auto-allo). The response rate after ASCT was 86% and 34 patients (52% of responding patients) obtained complete remission before allogeneic SCT. After a median follow up of 38 months after allogeneic stem cell transplantation, the 3-year overall survival,, progression-free survival, non-relapse mortality and relapse/progression were 68% (95%CI: 59-77), 61% (95%CI: 52-70), 17% (95%CI: 10-25) and 22% (95%CI: 14-30), respectively. In multivariate analysis, the response to autologous stem cell transplantation was the only independent predictive factor of mortality (P=0.05), whereas autologous stem cell transplantation conditioning and the type of allogeneic donor did not significantly affect survival (P=0.40 and P=0.68, respectively). No survival difference was observed between Hodgkin and non-Hodgkin lymphoma patients (P=0.53).

Salvage chemotherapy followed by high-dose therapy and autologous sem cell transplantation (ASCT) is recognized as the most effective strategy for relapsed or refractory Hodgkin lymphoma (HL) or aggressive non-Hodgkin lymphoma (NHL).[1,2] However, patients with some adverse prognostic features still have unsatisfactory outcomes after ASCT alone and may benefit from additional therapies. Short response duration after first-line therapy, B symptoms, advanced stage and/or extranodal disease at relapse and relapse in a previously irradiated field are among the most recognized adverse prognostic factors in relapsed/refractory HL patients,[3,4] whereas relapse within one year of diagnosis, prior rituximab therapy and secondary IPI score have been reported in diffuse large B-cell lymphoma.[5] Other prognostic factors have been identified for relapsed follicular and mantle cell lymphoma. Despite the identification of several adverse prognostic factors, the question about the best therapy for patients with refractory lymphoma is still open, and efforts are ongoing in an attempt to investigate the role and the timing of high-dose chemotherapy, allogeneic SCT, and investigational drugs. The aim of the present retrospective analysis is to report data on the feasibility, toxicity and outcome of tandem auto-allo in 111 adults with high-risk HL or NHL. Since it is known that the graft-*versus*-lymphoma (GvL) effect after allogeneic SCT is more effective when the tumor burden is minimal or not detected before transplantation, we explored a tandem strategy combining cytoreduction (through the administration of HDC followed by ASCT) and the GvL effect in those high-risk patients for whom ASCT is expected to produce unsatisfactory results.

From June 2002 to September 2013, we identified 111 patients with a diagnosis of lymphoproliferative disease who received tandem auto-allo at any point during their therapeutic history at two institutions (Istituto Clinico Humanitas, Milan, Italy; Institut Paoli-Calmettes, Marseille, France), selected from the internal transplantation database. The data regarding patients' age, sex, diagnosis, indication for tandem auto-allo, type and number of previous therapy or therapies, ASCT conditioning, interval between ASCT and allogeneic SCT, donor type and HCT-CI score were collected and analyzed as covariates in regression analyses; the hematopoietic cell transplantation-specific comorbidity index was reported or retrospectively calculated whenever possible. First-line therapy was ABVD (HL patients), CHOP or CHOP-like regimens (patients with aggressive NHL, with the addition of rituximab in cases of malignant-cell positivity for CD20) or chemotherapy associations not including anthracyclines (i.e. R-CVP for indolent NHL). The salvage regimens were DHAP, IGEV or ICE with the addition of rituximab for $CD20^+$ lymphomas. As per institutional guidelines, BEAM or BEAM-like regimens were administered in Marseille whereas high-dose melphalan (HD-Mel) was administered in Milan. The allogeneic SCT conditioning regimens were classified as myeloablative (MAC), reduced intensity (RIC) or non-myeloablative (NMA) according to working definitions.[6] The choice of the conditioning depended on donor type, patient's clinical status, and any institutional protocols at the moment of patient enrollment. Regarding donor selection, when a patient lacked a suitable HLA-identical sibling, a search for a 10/10-matched or a 9/10-mismatched unrelated donor was initiated (only for patients aged ≤ 65 years at Istituto Clinico Humanitas). Beginning in 2010, in the absence of either an available HLA-identical sibling or an unrelated donor at the appropriate time interval, a haploidentical or cord blood donor was identified. Peripheral blood stem cells was the preferred source from HLA-identical and mismatched unrelated donors whereas haploidentical donors were scheduled to undergo bone marrow harvest under general anesthesia unless contraindicated. Response was assessed using the standard and revised Cheson criteria, these latter after the introduction of PET as a tool for response evaluation, occurring in 2003 and progressively used for disease evaluation of HL and aggressive NHL. The results pertaining to some patients in this cohort have been published elsewhere.[7,8] Patients were followed-up until June 2014. The study was approved by the IRB of both institutions.

The main patients' and transplants' characteristics are shown in Table 1. Ten patients were allocated to tandem auto-allo at diagnosis (n=3 mantle cell, n=3 transformed follicular, n=3 peripheral T-cell, n=1 NK lymphoma), based on disease aggressiveness, patient's physical status and willingness, clinical judgment. The indication of tandem auto-allo was given before ASCT for all patients and did not depend on response after ASCT (Table 1). The median interval between ASCT and allogeneic SCT was 85 days (range: 36-235). After ASCT, median neutrophil engraftment occurred at day 13 (range: 8-41). NCI-CTC grade 3-4 mucositis occurred in 49 patients (44%) and at least one moderate to severe infectious disease was documented in 34 patients (31%), without significant differences between (B)EAM and HD-Mel (P=0.69 and P=0.21, respectively). After allogeneic SCT, median neutrophil engraftment occurred at day 18 (range 11-42), day 23 (range: 15-32) and day 14 (range: 7-26) after peripheral blood stem cells, bone marrow and cord blood, respectively. Two graft failures occurred in 2 patients after haploidentical transplantation. Three patients died before engraftment (2 haploidentical, 1 HLA-identical). Grade 3-4 mucositis occurred in 7 patients (2 after MAC, 5 after RIC). The rate of moderate to severe infections during hospitalization was 0%, 15% and 26% after MAC, RIC and NMA conditioning, respectively. At the time of

ASCT, 46 patients (41%) were in complete remission (CR), 39 (35%) were in partial remission (PR), and 10 (9%) and 16 (15%) had stable (SD) or progressive disease (PD), respectively. Among the 65 patients with measurable disease before ASCT, response (i.e. CR or PR) was observed in 56 patients (86% overall response) without significant difference between (B)EAM and HD-Mel (*P*=0.28). All but one patient in CR before ASCT maintained CR before allogeneic SCT. Disease response to ASCT according to the diagnosis of HL, aggressive NHL and indolent NHL is shown in Figure 1. After a median follow up of 38 months after allogeneic SCT, the 3-year OS and PFS of the entire cohort were 68% (95%CI: 59-77) and 61% (95%CI: 52-70), respectively. The overall incidence of grade 2-4 and grade 3-4 acute graft-*versus*-host disease (GvHD) and chronic GvHD were 28% (95%CI: 20-37), 11% (95%CI: 5-17) and 40% (95%CI: 31-50), respectively. At three years, cumulative incidence of NRM and relapse/progression after allogeneic SCT were 17% (95%CI: 10-25) and 22% (95%CI: 14-30), respectively. Survival of patients in CR before allogeneic SCT was superior to those with active disease and patients obtaining CR after ASCT had the same survival probability of those who were already in CR before ASCT (Figure 2A). The overall mortality hazard ratio (HR) was 0.40 (95%CI: 0.16-1.00) for responders who obtained CR *vs.* non-responders (i.e. patients in SD or PD; *P*=0.05), whereas the HR of patients who converted to PR *versus* non-responders was 0.67 (95%CI: 0.24-1.90; *P*=0.46). In multivariate analysis, response to ASCT was the only independent predictive factor of OS. The survival advantage of patients in CR *versus* those not in CR was maintained across the three histology groups cited above (*P*-value of interaction term: 0.97) (Figures 2B-D). No statistically significant variable was found in the multivariate models of OS or RM with the exception of a higher NRM risk after transplant with a haplo- *versus* HLA-identical sibling: HR = 3.55 (95%CI: 1.06-11.89; *P*=0.04). However, 4 of the 5 toxic deaths after haplo-transplants occurred in patients who presented with progressive disease before ASCT, indeed donor type was no longer statistically significant in the NRM model after the variable "disease status before ASCT" was added (*data not shown*). Overall mortality did not differ according to the intensity of conditioning regimen of allogeneic SCT: among 36 total deaths (36 of 111, 32%), one event occurred after the 3 MAC (33%), 22 after the 65 RIC (34%), 13 after the 43 NMA (30%).

To our knowledge, so far few reports have been published on tandem autologous-allogeneic transplantation in lymphoma.[7,9-11] In addition to a recently reported experience in HLA-identical setting,[7] Cohen *et al.* reported

Table 1. Main patients' and transplant characteristics.

Variable	Value	%
N.	111	100%
Pt. age (median)	44	range: 16-69
Gender		
M	66	59%
F	45	41%
Disease		
HL	44	40%
DLBCL	12	11%
FL	21	19%
Transf FL	9	8%
MCL	9	8%
MZL	1	1%
T lymph	13	12%
Gray zone	1	1%
NK lymphoma	1	1%
HCT-CI		
0	31	28%
1-2	27	24%
>2	36	33%
missing	17	15%
Indication for tandem auto-allo		
primary refractory	28	25%
no CR after salvage	43	39%
histology	10	9%
relapse after prior ASCT	6	5%
multiple relapses	24	22%
Prior therapy lines (median) 2	range: 0-7	
Prior radiotherapy	26	23%
ASCT conditioning		
BEAM	54	49%
EAM¶	8	7%
HD-Mel 100@	1	1%
HD-Mel 140	12	11%
HD-Mel 200	33	30%
other°	3	3%
Allogeneic SCT conditioning		
MAC*	3	3%
RIC§	65	58%
NMA#	43	39%
Allogeneic stem cell donor		
HLA-id sibling	62	56%
MUD 10/10	24	22%
MUD 9/10	2	2%
haplo	20	18%
cord blood§	3	3%
GvHD prophylaxis		
CsA	59	53%
CsA + MMF	19	17%
CsA + MTX	12	11%
FK + MMF + PT-Cy	14	12%
CsA + MMF + PT-Cy	6	5%
none	1	1%
Graft source		
PBSC	91	82%
BM	16	14%
cord blood	3	3%
BM + PBSC	1	1%

*Hodgkin lymphoma; DLBCL: diffuse large B-cell lymphoma; FL: follicular lymphoma; Transf FL: transformed follicular lymphoma; MCL: mantle-cell lymphoma; MZL: marginal zone lymphoma; T lymph: T-cell lymphoma (including both ALK+ and ALK- anaplastic large T-cell lymphoma, peripheral T-cell lymphoma), HCT-CI: hematopoietic cell transplantation-specific comorbidity index; CR: complete response; BEAM: association of BCNU-etoposide-cytarabine-melphalan; EAM: association of etoposide-cytarabine-melphalan; HD-Mel: melphalan at doses from 100 to 200 mg/m²; CsA: cyclosporin A; MMF: mycophenolate mofetil; MTX: methotrexate; FK: tacrolimus; PT-Cy: post-transplant high-dose cyclophosphamide; PBSC: peripheral blood stem cells; BM: bone marrow. §BCNU was not administered if patient's DLCO < 50. @Dose reductions may have been realized depending on patient's age, comorbidities, renal function; °Associations of: BCNU-thiotepa (n=1), etoposide-melphalan (n=1), BCNU-etoposide-cytarabine-cyclophosphamide (n=1); *MAC consists of an association of fludarabine-busulfan-ATG (n=3); §RIC include associations of: fludarabine-busulfan-ATG (n=54), thiotepa-fludarabine-cyclophosphamide +/- ATG (n=6), BEAM (n=2), fludarabine-treosulfan-ATG (n=1), thiotepa-fludarabine-ATG-rituximab-TBI 200 cGy (n=1), thiotepa-melphalan-cyclophosphamide-ATG (n=1); #NMA include associations of: fludarabine-cyclophosphamide-TBI 200 cGy (n=22), fludarabine-TBI 200 cGy (n=11), fludarabine-cyclophosphamide (n=9), fludarabine-melphalan-alemtuzumab-TBI 200 cGy (n=1); ¶two single and one double unit. NOTE: some proportions may exceed 100% due to rounding.*

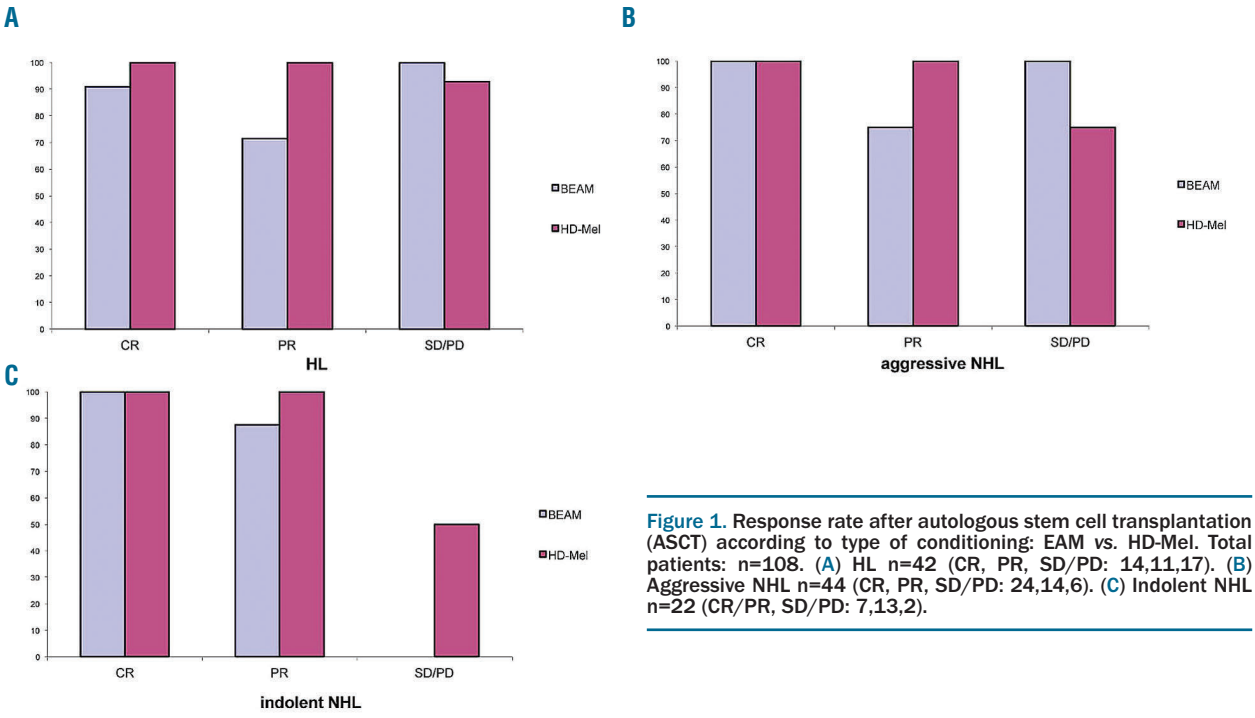

Figure 1. Response rate after autologous stem cell transplantation (ASCT) according to type of conditioning: EAM *vs.* HD-Mel. Total patients: n=108. (A) HL n=42 (CR, PR, SD/PD: 14,11,17). (B) Aggressive NHL n=44 (CR, PR, SD/PD: 24,14,6). (C) Indolent NHL n=22 (CR/PR, SD/PD: 7,13,2).

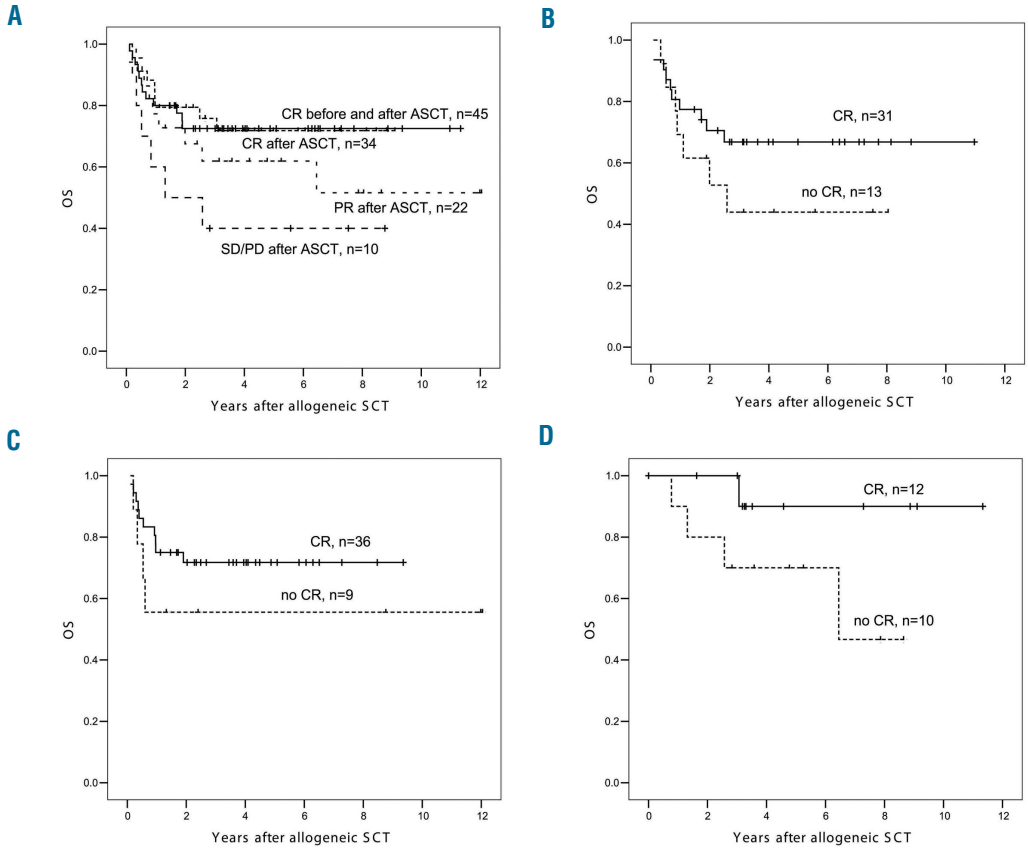

Figure 2 A-D. Overall survival (OS) and progression-free survival (PFS) according to disease status before allogeneic SCT. Estimates at three years for all patients. (A) OS: CR before and after autologous SCT (ASCT) (n=45), CR after ASCT (n=34), PR after ASCT (n=22), SD/PD after ASCT (n=10): 72% (95%CI: 59-85), 76% (95%CI: 61-91), 62% (95%CI: 42-82) and 40% (95%CI: 10-70), respectively; (B) OS according to CR *vs.* active disease before allogeneic SCT in HL patients (n=44): 67% (95%CI: 50-84) *vs.* 44% (95%CI: 16-72); (C) OS according to CR *vs.* active disease before allogeneic SCT in aggressive Hodgkin lymphoma (HL) patients (n=45): 72% (95%CI: 57-87) *vs.* 56% (95%CI: 23-89); (D) OS according to CR *vs.* active disease before allogeneic SCT in indolent HL patients (n=22): 90% (95%CI: 71-100) *vs.* 70% (95%CI: 42-98).

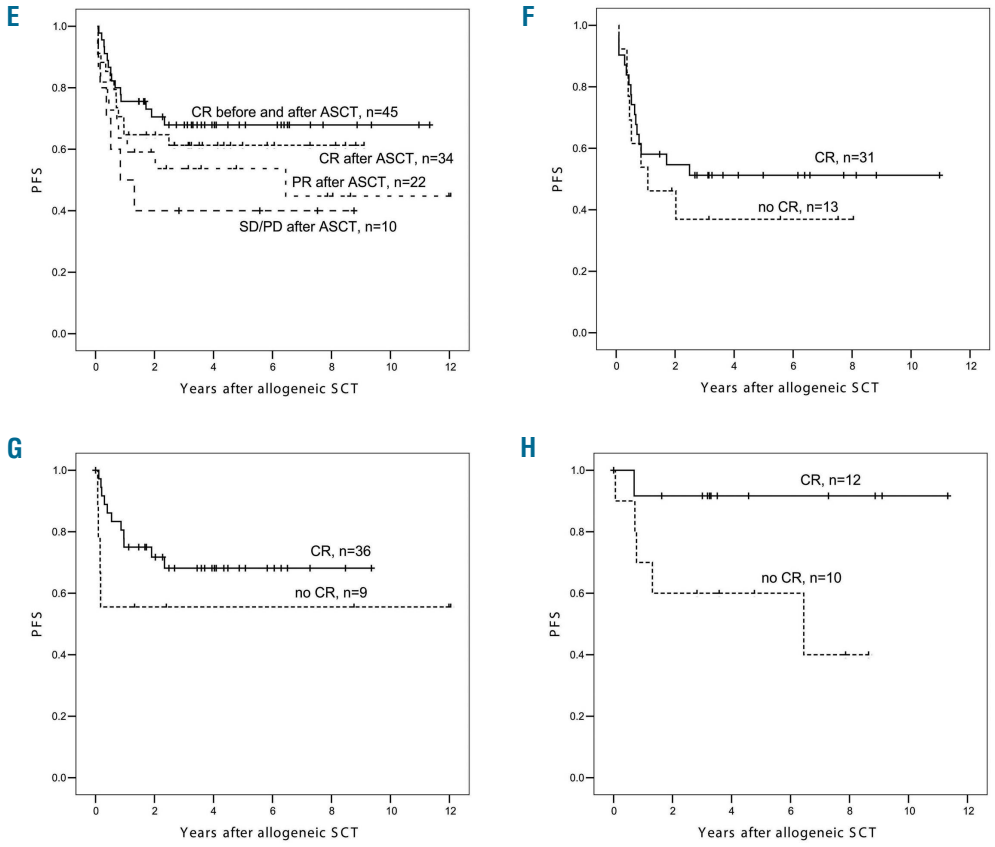

Figure 2 E-H (cont.). PFS: CR before and after ASCT (n=45), CR after ASCT (n=34), PR after ASCT (n=22), SD/PD after ASCT (n=10): 68% (95%CI: 54-82), 61% (95%CI: 45-77), 54% (95%CI: 34-74) and 40% (95%CI: 10-70), respectively; (F) PFS according to CR *vs.* active disease before allogeneic SCT in HL patients (n=44): 51% (95%CI: 33-69) *vs.* 37% (95%CI: 10-64); (G) PFS according to CR *vs.* active disease before allogeneic SCT in aggressive NHL patients (n=45): 68% (95%CI: 52-84) *vs.* 56% (95%CI: 23-89); (H) PFS according to CR *vs.* active disease before allogeneic SCT in indolent HL patients (n=22): 90% (95%CI: 71-100) *vs.* 60% (95%CI: 29-91).

excellent results in 27 patients with follicular lymphoma after allogeneic SCT from HLA-identical siblings with 96% OS and PFS at three years. Included in the present series were only 5 patients affected by follicular lymphoma who were transplanted by an HLA-identical donor after a fludarabine-cyclophosphamide-based conditioning; all are alive and disease-free at last follow up. A recent prospective study conducted on 30 pediatric patients affected by high-risk HL and NHL11 found similar outcomes compared with ours. Interestingly, the 3-year PFS of the 39 patients with aggressive NHL and chemosensitive disease (i.e. CR or PR before ASCT) was 65% (95%CI: 50-80) in our series; this finding appears to compare favorably with both the CORAL[5] and CIBMTR studies,[12] which reported a 3-year PFS of 53% and a 3-year OS of 50% in the same population, respectively. In particular, Gisselbrecht *et al.*[5] found a 39% PFS at three years in patients with unfavorable prognostic factors and who underwent ASCT. We believe that the GvL effect may have contributed to the fact that 2 of 3 of patients are alive and disease-free at three years from transplant in our cohort. Recently, Glass *et al.*[13] reported a promising 52% OS in a phase II trial with myeloablative allogeneic SCT in aggressive NHL patients, of whom 55% were classified as chemorefractory; the same study reported a 32% NRM, partially counterbalancing the beneficial GvL effect. A randomized trial would be of great interest in this subset of very high-risk patients. Another trial evaluating a myeloablative conditioning followed by allogeneic SCT (without prior ASCT) found an impressive 68% PFS among refractory HL patients.[14] The inferior outcome observed in our cohort may be explained at least in part by the slightly different population under investigation as 41% (18 of 44) of HL patients in our series were in SD or PD after salvage therapy *versus* 20% (5 of 25) in the UK trial.[14] An indirect role played by alemtuzumab against Reed-Stenberg cells could also be advocated.[15] Unexpectedly, we found no moderate or severe infections after MAC and more events after NMA than RIC; however, we believe that these data have to be interpreted with caution due to the fact that only 3 patients received MAC (all with an association of fludarabine-busulfan-ATG using 3 days of busulfan, namely reduced-toxicity conditioning) and that the difference between NMA and RIC is not statistically significant (P=0.22 by χ^2).

In conclusion, our experience with tandem autologous-allogeneic transplantation in 111 adult patients affected by high-risk HL and NHL suggests that this is a feasible and effective procedure for a selected population. Here, high-dose chemotherapy administered 2-3 months before allogeneic SCT provided a 86% response rate among patients who were not in CR before ASCT and, notably, those patients converting to CR had the same survival as those who were already in CR, thus underscoring the importance of further tumor shrinkage before

allogeneic immunotherapy in this subset of high-risk patients, irrespective of the conditioning intensity before allogeneic SCT. In our hands, BEAM and HD-Mel provided comparable results, both in terms of toxicity and efficacy against lymphoma.

Roberto Crocchiolo,[1] Luca Castagna,[1] Sylvain Garciaz,[2] Sabine Fürst,[2] Jean El Cheikh,[2] Barbara Sarina,[1] Stefania Bramanti,[1] Angela Granata,[2] Andrea Vai,[1] Samia Harbi,[2] Lucio Morabito,[1] Bilal Mohty,[2] Laura Giordano,[3] Raynier Devillier,[2] Diane Coso,[2] Monica Balzarotti,[1] Christian Chabannon,[2] Carmelo Carlo-Stella,[1] Armando Santoro,[1] Reda Bouabdallah[2] and Didier Blaise[2]

[1]Hematology Unit, Humanitas Clinical and Research Center, Rozzano, Italy; [2]Hematology Department, Institut Paoli-Calmettes, Marseille, France; and [3]Biostatistic Unit, Humanitas Clinical and Research Center, Rozzano, Italy

Correspondence: roberto.crocchiolo@humanitas.it
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References

1. Linch DC, Winfield D, Goldstone AH, et al. Dose intensification with autologous bone-marrow transplantation in relapsed and resistant Hodgkin's disease: results of a BNLI randomised trial. Lancet. 1993;341(8852):1051-1054.
2. Philip T, Guglielmi C, Hagenbeek A, et al. Autologous bone marrow transplantation as compared with salvage chemotherapy in relapses of chemotherapy-sensitive non-Hodgkin's lymphoma. N Engl J Med. 1995;333(23):1540-1545.
3. Lohri A, Barnett M, Fairey RN, et al. Outcome of treatment of first relapse of Hodgkin's disease after primary chemotherapy: identification of risk factors from the British Columbia experience 1970 to 1988. Blood. 1991;77(10):2292-2298.
4. Brice P, Bastion Y, Divine M, et al. Analysis of prognostic factors after the first relapse of Hodgkin's disease in 187 patients. Cancer. 1996; 78(6):1293-1299.
5. Gisselbrecht C, Glass B, Mounier N, et al. Salvage regimens with autologous transplantation for relapsed large B-cell lymphoma in the rituximab era. J Clin Oncol 2010;28(27):4184-4190.
6. Bacigalupo A, Ballen K, Rizzo D, et al. Defining the intensity of conditioning regimens: working definitions. Biol Blood Marrow Transplant. 2009;15(12):1628-1633.
7. Crocchiolo R, Castagna L, Fürst S, et al. Tandem autologous-allo-SCT is feasible in patients with high-risk relapsed non-Hodgkin's lymphoma. Bone Marrow Transplant. 2013;48(2):249-252.
8. Castagna L, Crocchiolo R, Giordano L, et al. High dose melphalan with autologous stem cell support in FDG PET-refractory Hodgkin lymphoma patients as bridge to second transplant. Bone Marrow Transplant. 2015;50(4):499-504.
9. Carella AM, Cavaliere M, Lerma E, et al. Autografting followed by nonmyeloablative immunosuppressive chemotherapy and allogeneic peripheral-blood hematopoietic stem-cell transplantation as treatment of resistant Hodgkin's disease and non-Hodgkin's lymphoma. J Clin Oncol. 2000;18(23):3918-3924.
10. Cohen S, Kiss T, Lachance S, et al. Tandem autologous-allogeneic nonmyeloablative sibling transplantation in relapsed follicular lymphoma leads to impressive progression-free survival with minimal toxicity. Biol Blood Marrow Transplant. 2012;18(6):951-957.
11. Satwani P, Jin Z, Martin PL, et al. Sequential myeloablative autologous stem cell transplantation and reduced intensity allogeneic hematopoietic cell transplantation is safe and feasible in children, adolescents and young adults with poor-risk refractory or recurrent Hodgkin and non-Hodgkin lymphoma. Leukemia 2015;29(2):448-455.
12. Hamadani M, Hari PN, Zhang Y, et al. Early failure of frontline rituximab.containing chemo-immunotherapy in diffuse large B cell lymphoma dose not predict futility of autologous hematopoietic cell transplantation. Biol Blood Marrow Transplant. 2014;20(11):1729-1736.
13. Glass B, Hasenkamp J, Wulf G, et al. Rituximab after lymphoma-directed conditioning and allogeneic stem-cell transplantation for relapsed and refractory aggressive non-Hodgkin lymphoma (DSHNHL R3): an open-label, randomised, phase 2 trial. Lancet Oncol. 2014;15(7):757-766.
14. Thomson KJ, Kayani I, Ardeshna K, et al. A response-adjusted PET-based transplantation strategy in primary resistant and relapsed Hodgkin Lymphoma. Leukemia. 2013;27(6):1419-1422.
15. Peggs KS, Sureda A, Qian W, et al. Reduced-intensity conditioning for allogeneic haematopoietic stem cell transplantation in relapsed and refractory Hodgkin lymphoma: impact of alemtuzumab and donor lymphocyte infusions on long-term outcomes. Br J Haematol. 2007;139(1):70-80.