Allogeneic stem cell transplantation for advanced acute promyelocytic leukemia in the ATRA and ATO era

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ABSTRACT

The role of allogeneic stem cell transplant in advanced acute promyelocytic leukemia patients who received standard first- and second-line therapy is still unknown. We report the outcome of 31 acute promyelocytic leukemia patients (median age 39 years) who underwent allogeneic transplant in second remission (n=15) or beyond (n=16). Sixteen patients were real-time polymerase chain reaction positive and 15 negative for *PML/RARA* pre-transplant. The 4-year overall survival was 62% and 31% for patients transplanted in second remission and beyond, respectively (P=0.05), and 64% and 27% for patients with pre-transplant negative and positive real-time polymerase chain reaction, respectively (P=0.03). The 4-year cumulative incidence of relapse was 32% and 44% for patients transplanted in second remission and beyond, respectively (P=0.37), and 30% and 47% for patients transplanted with negative and positive real-time polymerase chain reaction, respectively (P=0.30). Transplant-related mortality was 19.6%. In conclusion, allogeneic transplant is effective in advanced acute promyelocytic leukemia in the all-trans-retinoic acid and arsenic trioxide era, and should be considered once relapse is diagnosed.

Key words: allogeneic stem cell transplant, acute promyelocytic leukemia, acute promyelocytic leukemia relapse, all-trans retinoic acid, arsenic trioxide.

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Introduction

Approximately 80% of patients with acute promyelocytic leukemia (APL) are cured with modern all-trans-retinoic acid (ATRA) and chemotherapy.[1] As for patients who relapse after this regimen, arsenic trioxide (ATO) with or without other agents is considered the treatment of choice as it produces second molecular remission rates of up to 80%.[2,3] Available options for consolidation after remission re-induction include continued ATO and/or chemotherapy plus ATRA, with or without autologous or allogeneic transplants.[1] However, the best option for consolidation therapy for APL in second complete remission (CR2) is still unknown. Selection of the appropriate consolidation depends on variables which include patient age and performance status, duration of first remission, donor availability, and minimal residual disease status after salvage therapy, as determined by nested real-time polymerase chain reaction (RT-PCR) or quantitative PCR of the *PML/RARA* fusion gene.[1,3] Due to the very low number of relapses and the above variables, appropriately designed randomized studies to compare the different consolidation approaches in CR2 are deemed not feasible.

While the graft-*versus*-leukemia effect of allogeneic transplant offers the best anti-leukemia activity in patients with advanced disease, its success is notoriously hampered by transplant-related mortality which increases considerably in the matched unrelated and haploidentical settings.[4,5] Therefore, allogeneic transplant in APL is generally recommended for patients failing to achieve first or second molecular remission, or in those experiencing short duration of first remission.[1,3]

To date, four major studies have explored the role of allogeneic transplants in adult APL patients in the ATRA era.[4,6-8] We previously reported a single center study on the outcome of allogeneic transplant in 17 relapsed APL patients for whom pre-transplant minimal residual disease (MRD) assessment was available. We documented a significant anti-leukemic effect of allogeneic transplant even in patients with advanced disease including those with pre-transplant evidence of MRD.[6] A survey conducted by the European Bone Marrow Transplant on a large number of cases showed that allogeneic transplant significantly reduced relapse rate compared to autologous transplants. However, this benefit was at the expense of increased transplant-related mortality that

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reached 24±5%.[4] Major limitations of this large survey were the lack of information on pre-transplant treatments and status of MRD prior to transplant. None of the previous studies specifically focused on the role of allogeneic transplant after the advent of arsenic trioxide. Here we report on results of transplant in advanced APL patients treated in the ATRA and ATO era whose diseases were monitored by RT-PCR prior to and after transplant.

Design and Methods

Pre-transplant and transplant characteristics

Data on 31 APL patients (median age 39 years, range 16-55 years) with genetically confirmed APL who received allogeneic transplants were collected from 5 Italian transplant centers (Universities of Tor-Vergata and La Sapienza of Rome, University of Bologna, University of Perugia, and San Martino Hospital, Genoa). All patients with APL undergoing allogeneic transplants in these institutions between July 2000 and July 2010 were included. None of these patients had been included in our previous report.[6] All protocols received approval from the corresponding institutional review board and/or ethics committees. Patients were treated accordingly after signing informed consent. Reverse-transcriptase polymerase chain reaction identification and monitoring of the *PML/RARA* hybrid gene was carried out at local institutions following the standardized procedures described by the Biomed-I concerted action.[9] In all cases, post-transplant relapses were confirmed by RT-PCR amplification of the *PML/RARA* hybrid gene. Main patients' characteristics at first diagnosis, treatment prior to transplant, pre-transplant RT-PCR status and transplant characteristics are summarized in Table 1 and in the *Online Supplementary Table S1*. Information concerning first-line and salvage therapy was available for 31 and 29 patients. As first-line therapy, 29 patients received the AIDA regimen (ATRA plus idarubicin) while one patient (UPN15) received idarubicin, cytarabine and etoposide, and the other (UPN21) received DNR and Ara-C. Median duration of CR1 was 19.5 months (range 6-60 months). Seventeen patients received ATO as a salvage therapy (11 as second-line, 5 as third-line and one as fourth-line therapy). The remaining 12 patients received at relapse ATRA plus chemotherapy or gemtuzumab ozogamicin (GO, n=5 patients), chemotherapy alone (n=6) or single agent GO (n=1).

Fifteen patients received transplants in CR2 and 16 in CR3 or beyond (Table 1). Median time from remission to allogeneic transplant was five months (range 1-18 months). At the time of transplant, 15 patients were in molecular and 16 in hematologic complete remissions with RT-PCR-positive for *PML/RARA* (*Online Supplementary Table S1*). The type of transplant, conditioning regimens, graft-*versus*-host-disease (GvHD) prophylaxis, and status at last follow up are shown in Table 1. Eighteen patients were transplanted from HLA identical siblings (Id-sib), 6 from matched unrelated donors (MUD) and the remaining 7 patients received haploidentical transplant. Peripheral blood and bone marrow were the sources in 18 and 13 patients, respectively. Thirty patients received myeloablative conditioning regimens which were TBI-based in 15 and chemotherapy-based in 15 patients. One patient (UPN9) received reduced intensity conditioning regimen. Graft-versus-host-disease prophylaxis included cyclosporine A with a short course of methotrexate (26 patients), together with antithymocyte globulin (n=2), and antithymocyte globulin (ATG) alone (n=3).

End points and statistical analysis

Median follow up of the whole series was 55 months (range 4-100 months). Graft-*versus*-host-disease (GvHD) was graded as described elsewhere.[10] Unadjusted time-to-event analyses were performed using the Kaplan-Meier estimate. Overall survival (OS) was calculated from the transplant date to death or last follow up, cumulative incidence of relapse (CIR) was calculated from the date of transplant to relapse (hematologic or molecular), persistence of PCR-positivity post-transplant or to date of last follow up for patients alive in molecular remission, using the cumulative incidence method and considering death in remission as competing risk. The log rank test was used to compare risk factor categories for the Kaplan-Meier and the cumulative incidence curves. Transplant-related mortality was defined as all causes of non-leukemic deaths. Kaplan Meier curves and log-rank analysis were performed using the R-2.11.0 software program.

Results and Discussion

Fourteen of 31 patients (45%) are currently alive in complete remission at a median follow up of 55 months (range 4-100 months) after transplant. A total of 13 patients relapsed after transplant, of whom 11 died of disease progression at a median of eight months (range 3-59 months). The 2 remaining patients, one with molecular and the other with hematologic and CNS relapse, achieved a new molecular remission after ATO plus ATRA and tamibarotene followed by ATO and radiotherapy, respectively. Non-leukemia death was reported in 6 cases: GvHD grade IV, n=2 patients; multiorgan failure, n=1; cytomegalovirus (CMV) pneumonia, n=1; unspecified infection, n=2. Four-year cumulative incidence of transplant-related mortality was 19.6%.

A total of 15 patients developed acute GvHD: liver grade III-IV, n=4; skin grade I-II, n=10; skin grade III, n=1. Chronic GvHD occurred in 7 patients: limited skin, n=3; extensive skin, n=1; intestinal GvHD, n=3.

The probability of survival at four years was 45% for the whole study group, 62% and 31% for patients transplanted at CR2 and CR3 or over, respectively (P=0.05, Figure 1A). There was a significant difference in the 4-year OS between patients transplanted in molecular remission and those transplanted in hematologic remission but RT-PCR-positive at time of transplant (64% and 27%, respectively (P=0.03, Figure 1B). The 4-year CIR was 38% for all patients, 32% and 44% for patients transplanted at CR2 and CR3 or over, respectively (P=0.37, Figure 2A). Similarly, the 4-year CIR was lower in patients transplanted while in molecular remission compared to those in remission but with RT-PCR-positive disease at transplant (30% and 47%, respectively; P=0.30, Figure 2B). We acknowledge that compared to previous reports on allogeneic transplants in APL, our study includes a limited series from selected institutions. However, we provide detailed information on molecular disease status pre-transplant which is lacking in the majority of larger studies reported to date.[4] This information allows a much better evaluation of transplant efficacy, particularly in patients with persistent molecular disease and/or in advanced phase who had previously received state-of-art therapy. All but 2 patients in our series had received the AIDA protocol as front-line and more than half received ATO as a salvage therapy. Furthermore, 24 of the 31 cases had very advanced disease and poor prognostic features (i.e. 16 patients transplanted in CR3 or over and 8 patients in CR2 but RT-PCR-positive at transplant).

Previous studies have reported a lower relapse rate in

Table 1. Transplant features and outcome.

UPN	Transplant type/ source	Conditioning regimen	GVHD prophylaxis	Disease status at SCT	*PML/RARA* RT-PCR at SCT	Status at last FU (month)	Cause of death
UPN1	MUD/BM	BU based	CSA+MTX	CR2	negative	CR (20)	NA
UPN2	HLAI/BM	BU based	CSA+MTX	CR2	negative	Dead (59)	Disease relapse
UPN3	HLAI/BM	BU based	CSA+MTX	CR2	negative	Dead (26)	Disease relapse
UPN4	HLAI/BM	BU based	CSA+MTX	CR2	negative	CR (61)	NA
UPN5	HLAI/BM	BU based	CSA+MTX	CR2	negative	CR (55)	NA
UPN6	HLAI/BM	BU based	CSA+MTX	CR2	negative	CR (43)	NA
UPN7	Haplo/PBSC	TBI-based	CSA+ATG	CR2	negative	CR (7) lost FU	NA
UPN8	HLAI/PBSC	BU based	CSA+MTX	CR3	negative	CR (66)	NA
UPN9	HLAI/PBSC	FLU-AraC-ATG	CSA+MTX	CR3	negative	Dead (12)	Disease relapse
UPN10	HLAI/PBSC	TBI-based	CSA+MTX	CR3	negative	CR (22)	NA
UPN11	MUD/PBSC	TBI-based	CSA+MTX	CR3	negative	Dead (7)	Disease relapse
UPN12	MUD/PBSC	TBI-based	CSA+MTX	CR3	negative	Dead (3.5)	TRM (GVHD)
UPN13	HLAI/PBSC	BU based	CSA+MTX	CR3	negative	CR (70)	NA
UPN14	Haplo/PBSC	TBI-based	ATG	CR3	negative	CR (62)	NA
UPN15	Haplo/PBSC	TBI-based	CSA+ATG	CR4	negative	Dead (4)	TRM (CMV pneumonia)
UPN16	MUD/BM	BU based	CSA+MTX	CR2	positive	Dead (0.7)	TRM (MOF)
UPN17	HLAI/PBSC	TBI-based	CSA+MTX	CR2	positive	CR (50)	NA
UPN18	MUD/BM	TBI-based	CSA+MTX	CR2	positive	Dead (6)	TRM (infection)
UPN19	HLAI/BM	TBI-based	CSA+MTX	CR2	positive	Dead (9)	Disease relapse
UPN20	HLAI/PBSC	FLU-TT-IDA	CSA+MTX	CR2	positive	Dead (7)	Disease relapse
UPN21	HLAI/BM	TBI-based	CSA+MTX	CR2	positive	CR (4) lost FU	NA
UPN22	Haplo/BM	TT-CTX	CSA+MTX	CR2	positive	CR (68)	NA
UPN23	Haplo/BM	TBI-based	CSA+MTX	CR2	positive	CR (58)	NA
UPN24	HLAI/PBSC	BU based	CSA+MTX	CR3	positive	Dead (0.5)	TRM (GVHD)
UPN25	HLAI/PBSC	BU based	CSA+MTX	CR3	positive	Dead (5.4)	Disease relapse
UPN26	HLAI/PBSC	BU based	CSA+MTX	CR3	positive	Dead (13)	Disease relapse
UPN27	HLAI/BM	TT-CTX	CSA+MTX	CR3	positive	Dead (5)	TRM (infection)
UPN28	MUD/PBSC	TBI-based	CSA+MTX	CR3	positive	Dead (12)	Disease relapse
UPN29	Haplo/PBSC	TBI-based	ATG	CR4	positive	CR(100)	NA
UPN30	HLAI/PBSC	TBI-based	CSA+MTX	CR4	positive	Dead (3)	Disease relapse
UPN31	Haplo/PBSC	TBI-based	ATG	CR4	positive	Dead (5)	Disease relapse

MUD: matched unrelated donor; BM: bone marrow; HLAI: HLA identical; PBSC: peripheral blood stem cell; haplo: haploidentical; BU: busulphan-based; TBI: total body irradiation-based; FLU: fludarabine; AraC: cytarabine; TT: thiotepa, CTX: cyclophosphamide; ATG: antithymocyte globulin; IDA: idarubicin; CSA: cyclosporine A; MTX: methotrexate; SCT: stem cell transplant; FU: follow up; TRM: transplant-related mortality; GVHD: graft versus *host disease; MOF: multiorgan failure; CMV: cytomegalovirus.*

patients receiving allogeneic transplants compared to those given autologous transplants in CR2. These results were, however, counterbalanced by a high transplant-related mortality rate of up to 40%.[4,7,8,11] Here, we report a 4-year cumulative incidence of mortality of 19.6% in a series that included 7 haploidentical transplants. This improvement may reflect recent advances in transplant supportive measures, a wider use of peripheral blood stem cells, as well as better haploidentical transplant protocols.[4,5,12] In this context, we observed higher transplant-related mortality in patients transplanted before 2006 (4 of 14 patients; one transplanted in CR2, 3 in CR3 or over) than in patients transplanted from 2006 to 2010 (2 of 17; one transplanted in CR2, one in CR3 or over). Furthermore, in the current series, none of the 7 patients who underwent haploidentical transplants died as a result of transplant-related complications. The main cause of death in this series was disease relapse (63% of all deaths). This may reflect the presence of more advanced disease in our cohort. Indeed, relapse and death after relapse occurred mainly in patients who underwent allogeneic transplants after their second relapses (death after relapse in 7 of 16 transplanted in CR3 or over, 4 out of 15 transplanted in CR2). In these advanced stages, especially in patients with positive MRD, prophylactic donor lymphocyte infusion may reduce subsequent relapse in the post-transplant phase; a point that requires further investigation.[13,14] Despite a cohort of very advanced disease, overall survival (45%) and disease free survival (50%, *data not shown*) are comparable to previous reports on patients transplanted in CR2.[6,8] Thus, allogeneic transplant is capable of producing long-term remissions in such advanced disease. Previous studies have documented the efficacy of allogeneic transplant in relapsed APL patients who have achieved a second CR regardless of their pre-transplant RT-PCR results for minimal residual disease.[6,8] We

observed a better outcome for patients transplanted in second molecular remission compared to patients transplanted after multiple relapses (even after achieving molecular remission) and with persistent MRD at transplant (*Online Supplementary Table S2*). This suggests that both the number of remissions and the molecular status at time of transplant are important determinants of patient outcome.

Allogeneic transplant continues to be the preferred consolidation option in relapsed APL patients who are eligible for this treatment approach. Hence, the search for a donor is recommended immediately after relapse is diagnosed. However, in selected patients who had a long first remission duration (more than three years) and achieved a second molecular remission, other non-trans-

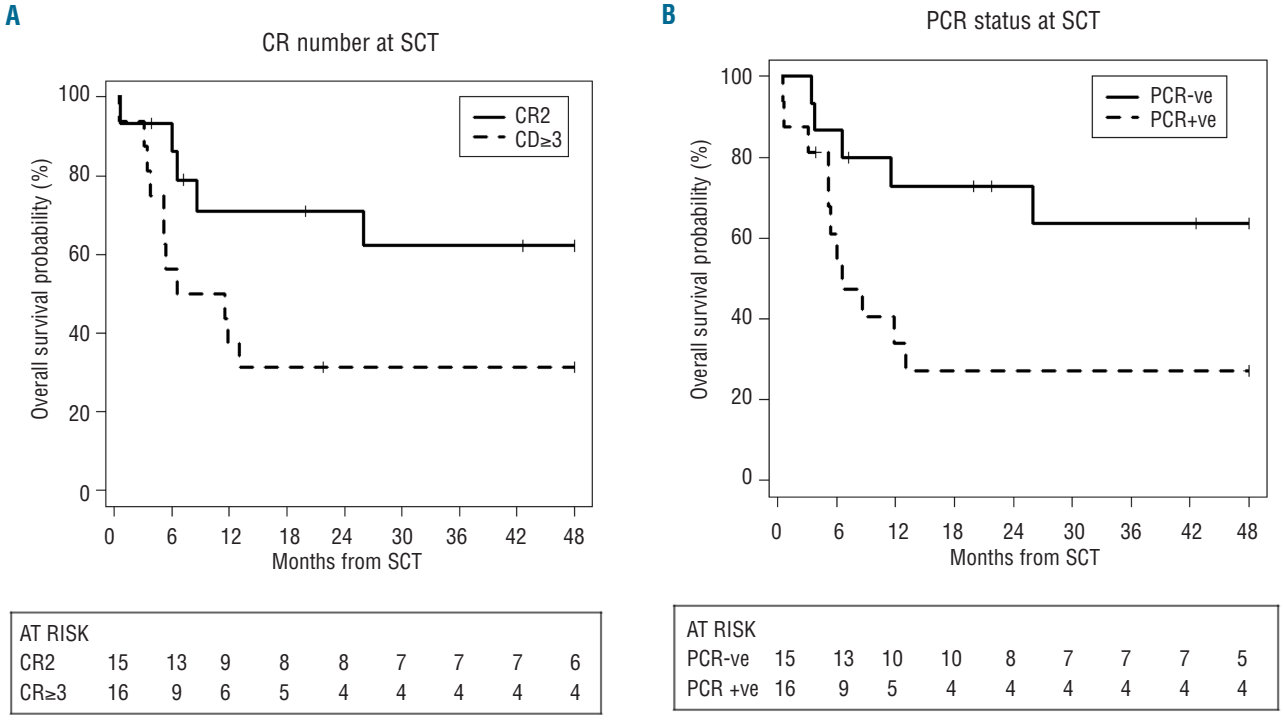

Figure 1. (A) **Overall survival for APL patients transplanted at second remission *versus* third remission or beyond.** (B) **Overall survival for APL patients transplanted with negative *versus* positive RT-PCR for *PML/RARA* at transplant.**

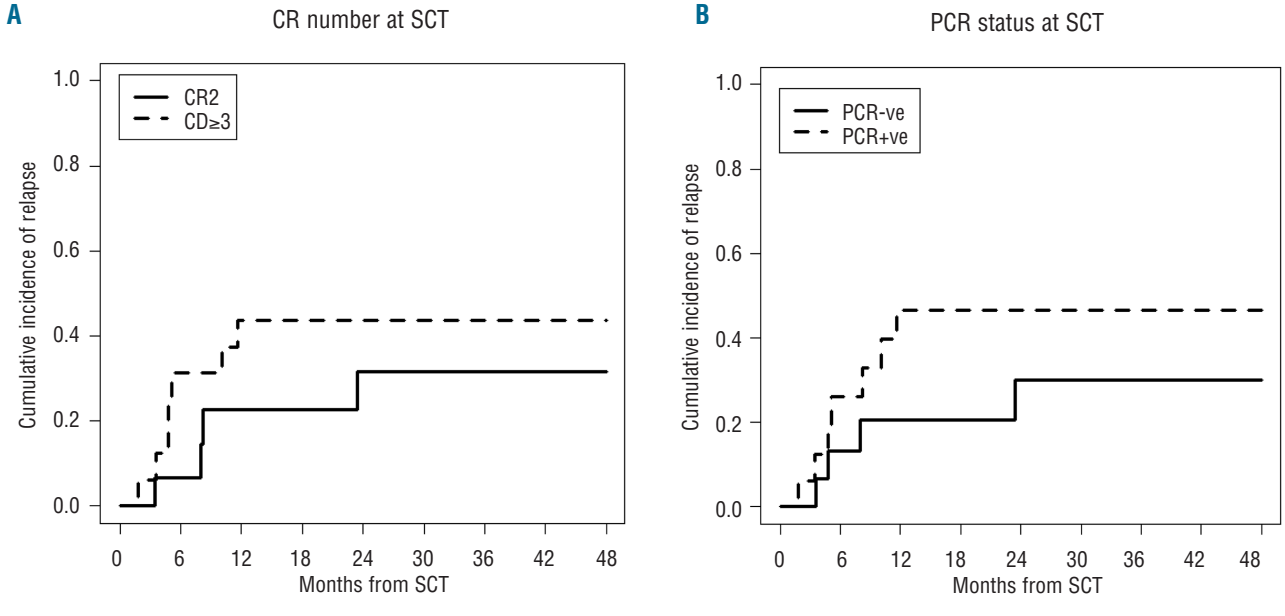

Figure 2. (A) **Cumulative incidence of relapse for APL patients transplanted in second remission versus third remission or beyond.** (B) **Cumulative incidence of relapse for APL patients transplanted with negative versus positive RT-PCR for PML/RARA at transplant.**

plant options such as prolonged arsenic trioxide or autologous transplant[15,16] can be a valid transplant alternative. The latter options should be compared to allogeneic transplant but the low relapse rate in APL would make prospective randomized trials impractical. It is worth emphasizing the efficacy and feasibility of allogeneic transplants in patients with more resistant or advanced disease.

Authorship and Disclosures

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