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ORIGINAL ARTICLE

Arsenic trioxide-based therapy of relapsed acute promyelocytic leukemia: registry results from the European LeukemiaNet

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In 2008, a European registry of relapsed acute promyelocytic leukemia was established by the European LeukemiaNet. Outcome data were available for 155 patients treated with arsenic trioxide in first relapse. In hematological relapse ($n = 104$), 91% of the patients entered complete hematological remission (CR), 7% had induction death and 2% resistance, 27% developed differentiation syndrome and 39% leukocytosis, whereas no death or side effects occurred in patients treated in molecular relapse ($n = 40$). The rate of molecular (m)CR was 74% in hematological and 62% in molecular relapse ($P = 0.3$). All patients with extramedullary relapse ($n = 11$) entered clinical and mCR. After 3.2 years median follow-up, the 3-year overall survival (OS) and cumulative incidence of second relapse were 68% and 41% in hematological relapse, 66% and 48% in molecular relapse and 90 and 11% in extramedullary relapse, respectively. After allogeneic or autologous transplantation in second CR ($n = 93$), the 3-year OS was 80% compared with 59% without transplantation ($n = 55$) ($P = 0.03$). Multivariable analysis demonstrated the favorable prognostic impact of first remission duration $\geqslant 1.5$ years, achievement of mCR and allogeneic or autologous transplantation on OS of patients alive after induction ($P = 0.03$, $P = 0.01$, $P = 0.01$) and on leukemia-free survival ($P = 0.006$, $P < 0.0001$, $P = 0.003$), respectively.

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INTRODUCTION

During the past decades, acute promyelocytic leukemia (APL) has developed from a highly fatal to the most curable type of acute myeloid leukemia.[1] Relapses after standard frontline therapy with all-trans retinoic acid (ATRA) and chemotherapy have become rare. Salvage regimens based on ATRA and chemotherapy induced high second remission rates but were no longer curative.[2–4] Subsequent autologous or allogeneic transplantation was a widely adopted strategy to consolidate second or later remissions, but the toxicity of the chemotherapy regimens led to a considerable rate of contraindications against transplantation and of fatal outcome.[4,5]

Arsenic trioxide (ATO) is presently regarded as the treatment of choice for relapsed APL after frontline therapy with ATRA and chemotherapy, yielding about 85% complete hematological remission (CR) rates, with limited toxicity, and particularly very limited myelosuppression.[6–8]

The optimal therapy to sustain ATO-induced remission is still unknown.[9] Although autologous transplantation appears to be curative only if performed in molecular remission, allogeneic transplantation may induce cure in patients with a positive reverse transcriptase–PCR (RT-PCR) for promyelocytic leukemia-retinoic acid receptor alpha (*PML-RARA*).[10,11] Whether other consolidation and/or maintenance strategies also have the potential for cure is disputed.[12,13]

It has also been suggested that treatment at the time of molecular relapse before overt hematological relapse improved the outcome of relapses,[14–16] but a clear benefit in long-term survival has not been proven so far.

Because of the rarity of relapses and different regulations in the European countries, a prospective clinical study could not be realized. Therefore, in 2008 an expert group established a European registry for relapsed APL to improve the information on the outcome with ATO. We report here the results of 155 patients treated with ATO in first relapse of APL.

METHODS

Objectives of the registry and eligibility

In 2008, a European registry of relapsed APL was established under the auspices of the European LeukemiaNet to gain insights into the clinical, biological and epidemiological characteristics of relapsed APL and to assess the curative potential of salvage therapy with ATO (PROMYSE Registry; registered at http://apps.who.int/trialsearch as DRKS00006761). Patients with genetically confirmed *PML-RARA*-positive first or successive molecular or clinical relapse of APL occurring from January 2003 onwards were eligible for retrospective or prospective registration independent of the administered therapy.

In prospectively registered patients, informed consent for inclusion in the registry was mandatory. Data of retrospectively registered APL patients were transferred from databases of national studies. In total, 237 patients have been included in this ongoing registry. This study focuses on the 155

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patients who received salvage therapy with ATO for first hematological, molecular or extramedullary relapse and were registered until the end of the year 2013.

Therapy of relapse

Treatment recommendations for relapsed APL were available at the website of the European LeukemiaNet (www.leukemia-net.org/content/) based on a consensus of the European APL Group of Experts.[17] Induction therapy was first recommended with ATO monotherapy, because an increased rate of APL differentiation syndrome by combination with ATRA was feared, but later ATO was frequently combined with ATRA. It was recommended to give a second course of ATO+ATRA as consolidation. After two ATO courses, an assessment of the molecular response by RT-PCR of *PML-RARA* was mandatory for further treatment decisions. Central nervous system (CNS) prophylaxis with intrathecal methotrexate was recommended.

Options for postconsolidation therapy were autologous and allogeneic stem cell transplantation, further ATO cycles or various modifications of maintenance therapy with or without ATO. The most appropriate treatment could be selected individually depending on several variables, including patient's age, performance status, PCR status, type of frontline therapy, first CR duration and donor availability. Of all the treatment options, only autologous transplantation was to be restricted to patients in molecular remission.[10] Supportive therapy and the management of APL differentiation syndrome and of leukocytosis were recommended according to international standards.[9]

Data management

Uniform online case record forms were used to ensure homogeneity in the documentation of data. In each country, a local coordinator was responsible for the registration of patients. The online database was centrally coordinated and evaluated at the University Hospital in Mannheim, Germany. The input of new data was screened regularly. Queries were sent to the local coordinators whenever relevant information was missing.

Definitions and statistical analysis

CR was defined according to Cheson *et al.*[18] and molecular remission by standardized nested RT-PCR or real-time quantitative PCR assays for *PML-RARA* performed in national reference laboratories.[14,19,20] Molecular persistence after two ATO cycles or relapse was confirmed by a repeat marrow aspirate taken within the next 2–4 weeks. Induction death was defined as death after start of therapy before achievement of CR. Overall survival (OS) was calculated from first relapse until death, and event-free survival from first relapse until non-achievement of remission, second relapse or death. Leukemia-free survival (LFS) was counted from the date of CR after first relapse until second molecular or hematological relapse or death in remission, whichever occurred first, and the cumulative incidence of relapse (CIR) from the date of CR until molecular or hematological relapse taking into account death in CR as competing risk.

The statistical analysis was performed using the SAS software (Version 9.2 for Windows, SAS Institute Inc., Cary, NC, USA). For categorical variables, the comparisons between hematological and molecular relapse were evaluated by Fisher's exact test and for continuous variables using the Mann–Whitney *U*-test. Distributions of time-to-event variables were estimated by the Kaplan–Meier method.[21] Comparisons were based on the log-rank test. CIRs were calculated according to Cooley.[22] Differences between the CIR curves were calculated with the Gray test.[23] All *P*-values reported are two sided.

Clinically important variables were analyzed concerning their prognostic influence, including gender, risk category at first diagnosis (Sanz's score low/intermediate versus high), duration of first remission $< / \geqslant 1.5$ years (which could better separate the groups than cutoffs at 1 or 2 years), type of relapse (hematological and molecular versus extramedullary), the application of ATRA during reinduction and/or consolidation (yes versus no) and of intensive chemotherapy for consolidation (yes versus no), the type of postconsolidation therapy (allogeneic or autologous transplantation versus no transplantation) and the RT-PCR status for *PML/RARA* after the end of consolidation (molecular remission versus no molecular remission). Univariable and multivariable analysis was performed using Cox proportional hazard model.[24] Factors with $P < 0.2$ in the univariable analysis were included in multivariable model with stepwise backward elimination.

RESULTS

Baseline patient characteristics

The 155 patients in first relapse of APL treated with ATO were registered in eight European countries (France 40, Germany 11, Greece 7, Italy 25, Spain 45, Sweden 1, Switzerland 4, United Kingdom 22 patients). They included 141 adults and 14 children or adolescents aged < 18 years. Twenty-two percent of the relapses occurred in the years 2003/2004, 26% in 2005/2006, 27% in 2006/2007 and 25% from 2009 to 2011. All relapses were genetically confirmed. Sanz's risk score at first diagnosis was low risk in 24%, intermediate risk in 47% and high risk in 29% of patients. All patients had received ATRA-plus-chemotherapy protocols used by APL cooperative groups in the different participating countries. No patient had received ATO during frontline therapy. As shown in Table 1, 104 (67%) patients had hematological, 40 (26%) molecular and 11 (7%) extramedullary relapse without hematological relapse ($n = 9$ in CNS, $n = 1$ vertebral/paravertebral mass, $n = 1$ cutaneous). In five of them, RT-PCR in the bone marrow was also positive. None of the patients diagnosed as molecular relapse had developed overt hematological relapse between diagnosis (marrow aspirate) and ATO onset (median 34 days, range 5 to 112).

The duration of first remission was shorter (median 512 days) in patients with molecular relapse than in patients with hematological relapse (median 764 days) ($P = 0.02$). Patients in hematological relapse had significantly lower median values of white blood cell count ($P = 0.04$), of platelet count ($P < 0.001$) and of hemoglobin ($P = 0.003$) and higher rates of bleeding ($P = 0.0008$) and coagulopathy ($P = 0.001$). Sanz's score at first diagnosis, the European Cooperative Oncology Group status and the PML/RARA isoform type did not differ significantly (Table 1).

Results of induction and consolidation therapy with ATO ± ATRA

The administered therapy in hematological, molecular and extramedullary relapse is shown in Table 2. For induction and consolidation course, the usual dosage of ATO was 0.15 mg/kg/day or regimens giving the same cumulative dose. The ATRA dose was uniformly 45 mg/m^2/day.

The treatment results are shown in Table 3. Ninety-five patients (91%) in hematological relapse achieved hematological CR (92 after induction and three after consolidation). Seven patients (7%) died during induction therapy and two (2%) remained resistant. Causes of death were CNS bleeding (on days 1, 3, 11, 19), infection (on day 32 and 64) or were unknown (on day 33). In patients with molecular or extramedullary relapse, no patient died during or after induction therapy.

After induction, the molecular remission rates in patients with hematological (53%) and molecular relapse (54%) were comparable ($P = 1.0$) (Table 3). The respective rates of molecular remission after consolidation were 74% and 62% ($P = 0.3$). The 11 patients (100%) with extramedullary relapse reached a molecular remission after consolidation therapy. The details of therapy and outcome of these patients are described in a separate section.

Results of postconsolidation therapy with or without transplantation

After ATO ± ATRA induction and consolidation treatment, patients underwent autologous ($n = 60$), allogeneic transplantation ($n = 33$) or received other treatments ($n = 55$) (Table 4). For autologous transplantation, the stem cell source was peripheral blood in almost all cases. The preferred conditioning regimen was busulfan combined with cyclophosphamide or melphalan. Approximately 80% of conditioning regimens before allogeneic transplantation were myeloablative (in the majority consisting of cyclophosphamide and total body irradiation) and 20% of reduced intensity.

Table 1. Patient characteristics at first relapse

	Hematological relapse			*Molecular relapse*			P-*value*[a]	*Extramedullary relapse*		
No of patients N = *155*		*104*			*40*				*11*	
Characteristics	*Median (range)*	*N*	*%*	*Median (range)*	*N*	*%*		*Median (range)*	*N*	*%*
Age at relapse (years)	42 (4–81)			46 (10–78)			0.61	46 (15–60)		
Male		70	67		25	63	0.69		6	55
Duration of first CR (days)	764 (31–3461)			512 (204–2362)			0.02	612 (166–2813)		
ECOG							0.27			
0–1		50/59	85		23/24	96			8/9	89
⩾2		9/59	15		1/24	4			1/9	11
WBC, $\times 10^9$/l	3.2 (0.5–112)			4.4 (1.9–7.6)			0.04	5.5 (2.1–9.0)		
Platelets, $\times 10^9$/l	66 (8–479)			187 (40–426)			< 0.001	157 (48–435)		
Hemoglobin, g/l	12.5 (5.5–17.2)			13.8 (9.1–16.1)			0.003	12.4 (8.7–15.7)		
PML/RARA isoform							0.31			
BCR1/BCR2		42/84	50		13/33	39			5/10	50
BCR3		42/84	50		20/33	61			5/10	50
Risk group at first diagnosis							0.46			
Low		28/103	27		8/40	20			1/11	9
Intermediate		49/103	48		18/40	45			5/11	45
High		26/103	25		14/40	35			5/11	45
Bleeding		21/90	23		0/33	0	0.0008		1/9	11
Coagulopathy		23/68	34		0/27	0	0.001		0/8	0

Abbreviations: CR, complete remission; ECOG, European Cooperative Oncology Group; PML, promyelocytic leukemia; RARA, retinoic acid receptor alpha; WBC, white blood cell. [a]*P*-value compares hematological and molecular relapse.

Table 2. Therapy of the patients with hematological, molecular or extramedullary relapse

	Hematological relapse			*Molecular relapse*			*P-value*[a]	*Extramedullary relapse*		
	Median (range)	N	*%*	*Median (range)*	N	*%*		*Median (range)*	N	*%*
No. of patients, N = 155		*104*			*40*				*11*	
Induction therapy							1.0			
ATO monotherapy		71/104	68		28/40	70			8/11	73
ATO+ATRA		33/104	32		12/40	30			3/11	27
Treatment duration of ATO ± ATRA (days)	31 (16–60)			29 (19–60)			0.21	27 (19–38)		
No. of patients, N = 148		*97*			*40*				*11*	
Consolidation therapy							0.006			
ATO monotherapy		45/74	61		13/36	36			1/10	10
ATO+ATRA		16/74	22		19/36	53			5/10	50
Systemic chemotherapy		13/74	17		4/36	19			4/10	40
No information		23			4				1	
Treatment duration of ATO ± ATRA (days)	25 (15–28)			25 (20–30)			0.9	25 (20–25)		
Intrathecal methotrexate		4/104	4		2/40	5	0.67		10/11	91
Postconsolidation therapy							0.34			
Autologous transplantation		42/97	43		12/40	30			6	55
Allogeneic transplantation		22/97	23		10/40	25			1	9
No transplantation in second CR		33/97	34		18/40	45			4	36

Abbreviations: ATRA, all-trans retinoic acid; ATO, arsenic trioxide; CR, complete remission. [a]*P*-value compares hematological and molecular relapse.

Table 3. Treatment results

	Hematological relapse		*Molecular relapse*		*P-value*[a]	*Extramedullary relapse*	
No. of patients N = 155	*104*		*40*			*11*	
	N	%	N	%		N	%
Results after induction							
CR (hematological)	92/104	88	–			11/11	100
Resistance (hematological)[b]	5/104	5	–			0	0
Death	7/104	7	0/40	0	0.19	0/11	0
Side effects of ATO during induction							
APL diff. syndrome	22/83	27	0/40	0	< 0.001	0/11	0
Leukocytosis[c]	36/92	39	0/40	0	< 0.001	0/11	0
Infection/FUO	27/63	43	3/29	10	0.002	4/11	36
Hepatotoxicity[d]	11/56	20	3/28	11	0.37	2/8	25
Rate of molecular remission							
After induction	40/76	53	21/39	54	1.0	9/9	100
After consolidation	39/53	74	18/29	62	0.32	11/11	100

Outcome	*% (95% CI)*	*% (95% CI)*		*% (95% CI)*
OS			0.85	
At 3 years	68 (58; 78)	66 (57; 75)		90 (82; 100)
EFS			0.57	
At 3 years	52 (41; 63)	45 (27; 63)		90 (71; 100)
No of patients N = 146	*95*	*40*		*11*
LFS			0.14	
At 3 years	55 (43; 68)	45 (27; 63)		80 (54; 100)
CIR			0.3	
At 3 years	41 (29; 52)	48 (29; 64)		11 (0; 42)

Abbreviations: APL diff. syndrome, acute promyelocytic leukemia differentiation syndrome; ATO, arsenic trioxide; CI, confidence interval; CIR, cumulative incidence of relapse; CR, complete remission; EFS, event free survival; FUO, fever of unknown origin; LFS, leukemia free survival; OS, overall survival. [a]*P*-value compares hematological and molecular relapse. [b]Three patients achieved remission after consolidation. [c]Leukocytosis requiring control with chemotherapy. [d]All grades of hepatoxicity.

Fifty-two percent of the patients who underwent allogeneic transplantation were RT-PCR positive for *PML/RARA* before transplantation compared with only 2% before autologous transplantation and with 17% of non-transplanted patients ($P < 0.001$). Non-transplanted patients were older ($P < 0.001$), but there was no significant difference between transplanted and non-transplanted patients for the Sanz risk group at first diagnosis ($P = 0.44$) and European Cooperative Oncology Group performance status at first relapse (0/1 versus $\geqslant 2$) ($P = 0.19$). There was also no significant difference in the proportion of allogeneic and autologous transplantation and non-transplant approaches between hematological and molecular relapses ($P = 0.34$).

In patients who were not transplanted ($n = 55$), information about postconsolidation therapy was available in 31 patients. In 21 of them, a variable number of additional ATO ± ATRA cycles were administered (median of 3). Eight patients received various chemotherapy regimes, one patient with simultaneous CNS relapse was irradiated and one patient received no further therapy. Twelve of those 55 patients underwent transplantation after second or subsequent relapse (allogeneic $n = 8$; autologous $n = 4$).

With a median follow-up of 3.2 years, 3-year OS and CIR were 70% and 42%, respectively, in the whole population (Figures 1a and b). The time of relapse (74 relapses occurred before the end of the year 2006 and 81 thereafter) had no prognostic influence (OS $P = 1.0$). The rate of death in CR was 4% (three deaths after infection and three not APL related). There was no difference in OS and CIR (OS: $P = 0.85$; and CIR: $P = 0.39$) between the patients in hematological and molecular relapse (Table 3 and Figures 2a and b). The results after autologous or allogeneic transplantation or other treatments are shown in Table 4 and Figures 3a and b. Furthermore, OS and CIR did not differ significantly between allogeneic and autologous transplantation (OS: $P = 0.57$; and CIR: $P = 0.7$). In the allogeneic group, OS was not significantly influenced by the PCR status before transplantation ($P = 0.14$). Three-year OS was 80% (95% CI (71; 89)) in the combined transplantation group (autologous or allogeneic) compared with 59% (95% CI (41; 76)) of non-transplanted patients ($P = 0.03$). The three-year CIR of the respective groups was 35% (95% CI (24; 47)) and 58% (95% CI (39; 77)) ($P = 0.02$).

Treatment of extramedullary relapses

All CNS relapses were treated with ATO ± ATRA-based systemic therapy and repeated intrathecal applications of methotrexate ± ara-C ± hydrocortisone, and all patients achieved clinical and molecular remission after induction or consolidation therapy (Table 3). Postremission therapy was heterogeneous. Three cases received partial or complete irradiation of the neuroaxis. Seven patients underwent autologous ($n = 6$) or allogeneic ($n = 1$) transplantation. One of these patients died in CR after infection. Non-transplanted patients ($n = 4$) received either irradiation

Table 4. Characteristics of patients who received autologous, allogeneic and no transplantation in second CR ($n = 148$)

	Autologous transplantation N = *60*		*Allogeneic transplantation* N = *33*		*No transplantation* N = *55*		P-*value*
	N	%	N	%	N	%	
PCR status							< 0.001
Negative	51/52	98	13/27	48	39/47	83	
Positive	1/52	2	14/27	52	8/47	17	
No information	8		6		8		
Age (years)							< 0.001
< 40	29/60	48	19/33	58	9/55	16	
40–59	28/60	47	14/33	42	21/55	38	
⩾ 60	3/60	5	0/33	0	25/55	46	
Risk group							0.44
Low	11/59	19	6/33	18	18/55	33	
Intermediate	30/59	51	17/33	52	22/55	40	
High	18/59	30	10/33	30	15/55	27	
ECOG							0.19
0–1	33/36	92	17/17	100	29/35	83	
2–3 (4)	3/36	8	0/17	0	6/35	17	
Outcome	*% (95% CI)*		*% (95% CI)*		*% (95% CI)*		
OS							0.09
At 3 years	77 (61; 94)		79 (67; 91)		59 (41; 76)		
CIR							0.05
At 3 years	37 (19; 56)		39 (24; 54)		59 (37; 76)		

Abbreviations: CI, confidence interval; CIR, cumulative incidence of relapse; CR, complete remission; ECOG, European Cooperative Oncology Group; OS, overall survival.

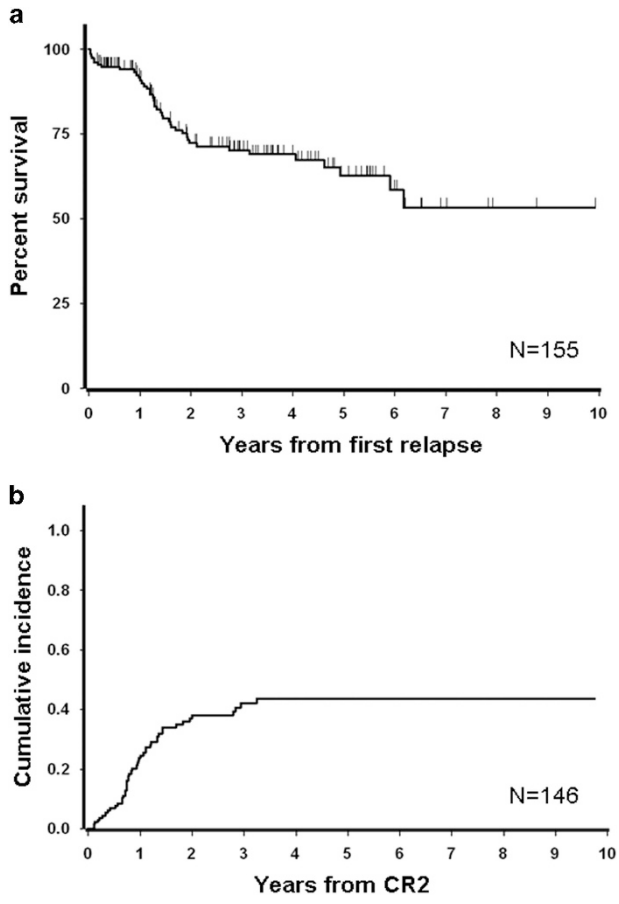

Figure 1. OS (**a**) and CIR (**b**) of all patients treated with ATO in first relapse of APL. In OS, tics indicate the last follow-up of the living patients.

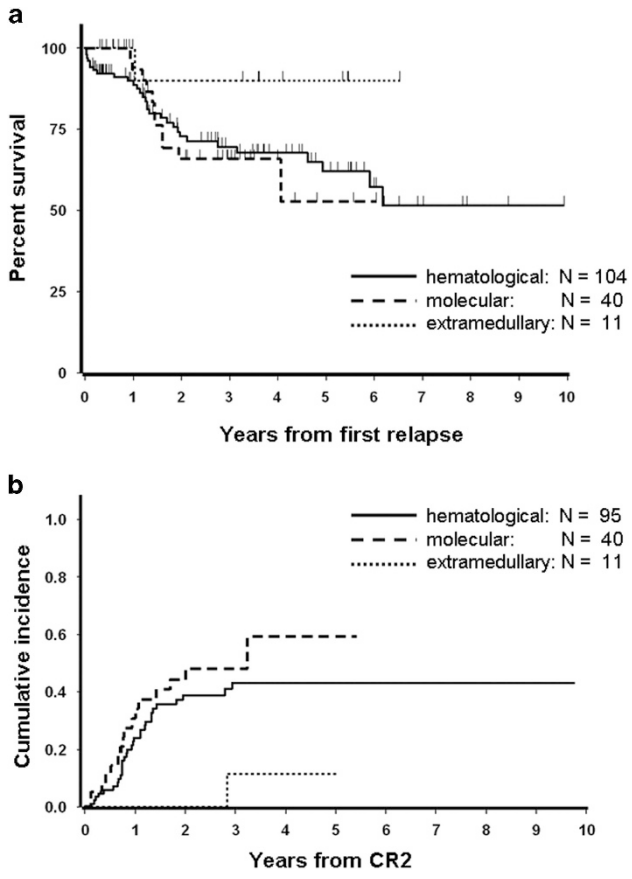

Figure 2. OS (**a**) and CIR (**b**) separated according to hematological, molecular or extramedullary relapse (OS $P = 0.31$; CIR $P = 0.047$, respectively). In OS, tics indicate the last follow-up of the living patients.

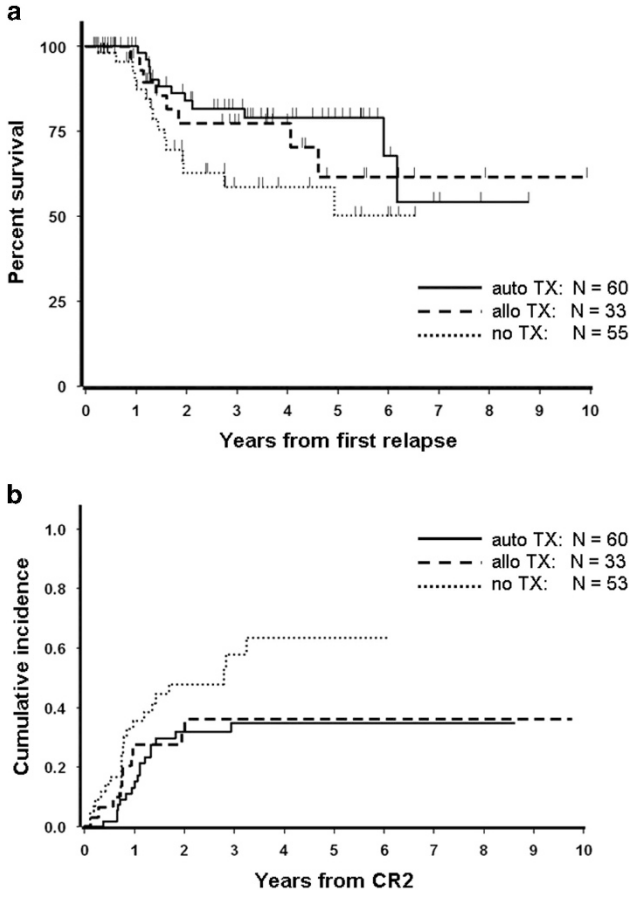

Figure 3. Outcome after postconsolidation therapy: OS (**a**) and CIR (**b**) of patients who underwent autologous or allogeneic transplantation or were not transplanted (OS $P = 0.09$; CIR $P = 0.05$, respectively). In OS, tics indicate the last follow-up of the living patients.

of the CNS ($n = 1$), regimens including high-dose ara-C ($n = 2$) or both ($n = 1$), and one patient relapsed. In the patient with promyelocytic sarcoma of the spine, a separate case report is available.[25] The patient with cutaneous promyelocytic sarcoma received induction therapy with ATO followed by consolidation with ara-C and autologous transplantation.

Side effects of induction treatment with ATO

Patients with hematological relapse had a significantly higher rate of ATO-related side effects during induction therapy than patients with molecular relapse (Table 3), including APL differentiation syndrome ($P < 0.001$), leukocytosis ($\geqslant 10 \times 10^9$/l) requiring control with chemotherapy (typically hydroxyurea) ($P < 0.001$) and rate of infections or fever of unknown origin ($P = 0.002$). The rate of hepatotoxicity (all grades) did not differ significantly. There were no evaluable data on QT-prolongation, as the measurements were not standardized.

Prognostic factors of outcome

Table 5 shows the results of univariable and multivariable analyses for OS of all patients and of the patients in hematological CR after consolidation and for LFS. The results of univariable and of multivariable analysis demonstrated a positive impact of first remission duration $\geqslant 1.5$ years, of achievement of second molecular remission and of transplantation in second CR (allogeneic or autologous) on OS of patients alive after ATO induction and on LFS.

DISCUSSION

To our knowledge, the present report includes the largest cohort of APL patients in first relapse of APL treated with ATO after frontline therapy with ATRA and chemotherapy. The results show that at least 50% of the patients in first relapse of APL can be cured by salvage therapy with ATO followed by consolidation therapy.

Table 5. Results of univariable and multivariable analysis of prognostic factors

	OS of all patients			*OS of patients alive after induction*			*LFS*		
Variable	*Univariable* P-*value*	*Multivariable HR (95% CI)*	P-*value*	*Univariable* P-*value*	*Multivariable HR (95% CI)*	P-*value*	*Univariable*	*Multivariable HR (95% CI)*	P-*value*
	N = *155*			N = *148*			N = *146*		
Gender (male versus female)	0.601			0.706			0.300		
Type of relapse[a]	0.161		0.173	0.209			0.103		0.20
WBC count ($\leqslant / > 10 \times 10^9$/l)	0.684			0.669			0.617		
Duration of first CR ($< / \geqslant 1.5$ years)	0.094		0.094	0.046	0.410 (0.183; 0.919)	0.03	0.004	0.401 (0.210; 0.764)	0.006
Additional ATRA (yes or no)				0.189		0.08	0.338		
Additional chemotherapy (yes or no)				0.294			0.328		
Tx versus no Tx				0.039	0.326 (0.139; 0.763)	0.01	0.017	0.363 (0.186; 0.708)	0.003
Molecular remission (yes or no)				0.019	0.314 (0.129; 0.764)	0.01	< 0.0001	0.249 (0.125; 0.496)	< 0.0001

Abbreviations: ATRA, all-trans retinoic acid; CI, confidence interval; CR, complete remission; HR, hazard ratio; LFS, leukemia-free survival; OS, overall survival; WBC, white blood cell. [a]Hematological and molecular relapse versus extramedullary relapse; Tx versus no Tx, allogeneic or autologous transplantation versus no transplantation in second CR.

Our data confirm the efficacy of ATO in reinduction of remission, as in 115 patients with hematological or extramedullary relapse, 91% entered CR and only 2% had resistant leukemia. These results are similar to the previously reported experiences with ATO, including a total of 304 patients with relapsed APL reported in >20 phase II studies, 86% of whom achieved hematological CR (reviewed in Lengfelder *et al.*[6]). In patients with molecular relapse, despite the lower leukemic burden compared with hematological relapse, the rates of molecular remission after induction (54% versus 53%) and consolidation therapy (62% versus 74%) did not differ significantly and were in agreement with previous reports.[6,7]

The rate of induction death, 5% and 7%, respectively in the overall series and in patients with hematological relapse was similar to previous experiences.[6] Of note, all induction deaths in our cohort occurred in patients with hematological relapse, mostly caused by fatal bleeding, and not in patients with molecular relapse. Patients in molecular relapse had more favorable clinical and laboratory parameters, which may explain the lower risk of bleeding and APL differentiation syndrome. On the other hand, and contrary to previous reports,[14,16,17,26] there was no survival advantage beyond 1 year in patients with molecular relapse, compared with those with hematological relapse, in spite of similar consolidation approaches. Nevertheless, the lower rate of early deaths and the fewer side effects of ATO support the importance of regular molecular monitoring to allow treatment of relapses at the molecular stage.

The OS of the 11 patients with extramedullary relapse was 90%. Nine of these patients had presented with a CNS relapse. Because of the variability of treatment after ATO, it is not possible to precisely assess the impact of the different therapeutic measures, including transplantation, in this small cohort. However, compared with the poor outcome previously associated with CNS relapse, it appears that ATO, which penetrates the CNS,[27] may contribute to improved prognosis.[28,29] Very recently, it was suggested that concomitant intravenous administration of mannitol could further enhance the CNS penetration of ATO.[30]

Analyses of autologous and allogeneic transplantation or non-transplant approaches for relapsed APL have been mostly retrospective and their results are heterogeneous, precluding the establishment of recommendations.[31–38] Our results indicate a potential survival benefit for transplantation (allogeneic or autologous transplantation) compared with no transplantation in second CR. It should be emphasized, however, that the comparability of the three patient groups is limited. Important differences are the high rate of molecular persistence before allogeneic transplantation compared with autologous transplantation (Table 4). Furthermore, the non-transplanted group included an older and heterogeneously treated population of patients who probably did not qualify for transplantation in the majority of cases. Considering these limitations, our results still support performing transplantation after first relapse, whenever possible. Our results also further indicate that patients at high risk of relapse, due to persistent PCR positivity, should undergo allogeneic transplantation, whereas patients in molecular remission probably benefit from autologous transplantation. Concerning the results without transplantation, the heterogeneity of the administered chemotherapy and the variability of the number of ATO cycles do not allow a clear assessment of these approaches. But interestingly, prolonged second remissions could be observed in those patients. These observations suggest that continuation of ATO might be an option in patients not qualifying for transplantation. Given the high effectiveness of expanded consolidation with four ATO courses in frontline therapy,[39] the application of more than one consolidation cycle might also be considered in relapsed patients assigned to transplantation, in particular if they have molecular persistence after the first consolidation course.

The 3-year OS of the 155 patients was 68%, suggesting a survival improvement after first relapse of approximately 20% with ATO-based salvage therapy by comparison to previous results obtained with ATRA and chemotherapy salvage.[8,40] Nevertheless, the relapse rate was still high (CIR 44% at 3 years) and uncontrolled leukemia after second or later relapse was the most frequent cause of death during follow-up. As shown by multivariable analysis, remission duration of <1.5 years, persistent PCR positivity after consolidation and the absence of transplantation were poor prognostic factors of OS and LFS (Table 5).

Limitations of this analysis are the retrospective nature of data collection and heterogeneous postconsolidation treatments applied. It should be emphasized, however, that virtually all patients had received state-of-the-art frontline therapy and that all participating countries followed the European treatment recommendations for relapsed APL,[17] thereby avoiding too large differences in the management of patients. Furthermore, the homogenous documentation of data was ensured by uniform case record forms. Under such conditions, registry data may provide valuable information on the outcome of patients with rare diseases, such as relapsed APL. The further registration of APL relapses after ATO+ATRA for frontline therapy[39] might allow to assess the outcome in this new era of APL therapy.

CONFLICT OF INTEREST

Eva Lengfelder and David Grimwade received research support from CEPHALON and TEVA. Francesco Lo-Coco and Maria Pagoni participated in speaker's bureau of TEVA. The other authors declare no conflict of interest.

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REFERENCES

1 Wang ZY, Chen Z. Acute promyelocytic leukemia: from highly fatal to highly curable. *Blood* 2008; **111**: 2505–2015.
2 Degos L, Dombret H, Chomienne C, Daniel MT, Micléa JM, Chastang C *et al.* All-trans-retinoic acid as a differentiating agent in the treatment of acute promyelocytic leukemia. *Blood* 1995; **85**: 2643–2653.
3 Castagnola C, Lunghi M, Corso A, Tajana M, Zappasodi P, Dabusti M *et al.* Management of acute promyelocytic leukemia relapse in the ATRA era. *Haematologica* 1998; **83**: 714–717.
4 Thomas X, Dombret H, Cordonnier C, Pigneux A, Gardin C, Guerci A *et al.* Treatment of relapsing acute promyelocytic leukemia by all-trans retinoic acid therapy followed by timed sequential chemotherapy and stem cell transplantation. *Leukemia* 2000; **14**: 1006–1013.
5 Sanz MA, Labopin M, Gorin NC, de la Rubia J, Arcese W, Meloni G *et al.* Hemotopoietic stem cell transplantation for adults with acute promyelocytic leukemia in the ATRA era: a survey of the European Cooperative Group for Blood and Marrow Transplantation. *Bone Marrow Transplant* 2007; **39**: 461–469.
6 Lengfelder E, Hofmann WK, Nowak D. Impact of arsenic trioxide in the treatment of acute promyelocytic leukemia. *Leukemia* 2012; **26**: 433–442.
7 Soignet SL, Frankel SR, Douer D, Tallman MS, Kantarjian H, Calleja E *et al.* United States multicenter study of arsenic trioxide in relapsed acute promyelocytic leukemia. *J Clin Oncol* 2001; **19**: 3852–3860.
8 Thomas X, Pigneux A, Raffoux E, Huguet F, Caillot D, Fenaux P. Superiority of an arsenic trioxide-based regimen over a historic control combining all-trans retinoic acid plus intensive chemotherapy in the treatment of relapsed acute promyelocytic leukemia. *Haematologica* 2006; **91**: 996–997.
9 Sanz MA, Grimwade D, Tallman MS, Lowenberg B, Fenaux P, Estey EH *et al.* Management of acute promyelocytic leukemia: recommendations from an expert panel on behalf of the European LeukemiaNet. *Blood* 2009; **113**: 1875–1891.
10 Meloni G, Diviero D, Vignetti M, Avvisati G, Capria S, Petti MC *et al.* Autologous bone marrow transplantation for acute promyelocytic leukemia in second remission: prognostic relevance of pretransplant minimal residual disease assessment by reverse transcription polymerase chain reaction of the PML/RARα fusion gene. *Blood* 1997; **90**: 1321–1325.

11 Ramadan SM, Di Veroli A, Camboni A, Breccia M, Iori AP, Aversa F *et al.* Allogeneic stem cell transplantation for advanced acute promyelocytic leukemia in the ATRA and ATO era. *Haematologica* 2012; **97**: 1731–1735.
12 Au WY, Lie AK, Chim CS, Liang R, Ma SK, Chan CH *et al.* Arsenic trioxide in comparison with chemotherapy and bone marrow transplantation for the treatment of relapsed acute promyelocytic leukaemia. *Ann Oncol* 2003; **14**: 752–757.
13 Lou Y, Qian W, Meng H, Mai W, Tong H, Tong Y *et al.* High efficacy of arsenic trioxide plus all-trans retinoic acid based induction and maintenance therapy in newly diagnosed acute promyelocytic leukemia. *Leuk Res* 2013; **37**: 37–42.
14 Grimwade D, Jovanovic JV, Hills RK, Nugent EA, Patel Y, Flora R *et al.* Prospective minimal residual disease monitoring to predict relapse of acute promyelocytic leukemia and to direct pre-emptive arsenic trioxide therapy. *J Clin Oncol* 2009; **27**: 3650–3658.
15 Lo Coco F, De Santis S, Esposito A, Divona M, Diverio D. Molecular monitoring of hematologic malignancies: current and future issues. *Semin Hematol* 2002; **39**: 14–17.
16 Esteve J, Escoda L, Martín G, Rubio V, Díaz-Mediavilla J, González M *et al.* Outcome of patients with acute promyelocytic leukemia failing to front-line treatment with all-trans retinoic acid and anthracycline-based chemotherapy (PETHEMA protocols LPA96 and LPA99): benefit of an early intervention. *Leukemia* 2007; **21**: 446–452.
17 Sanz MA, Fenaux P, Lo Coco FEuropean APL Group of Experts. Arsenic trioxide in the treatment of acute promyelocytic leukemia. A review of current evidence. *Haematologica* 2005; **90**: 1231–1235.
18 Cheson BD, Bennett JM, Kopecky KJ, Büchner T, Willman CL, Estey EH *et al.* Revised recommendations of the International Working Group for Diagnosis, Standardization of Response Criteria, Treatment Outcomes, and Reporting Standards for Therapeutic Trials in Acute Myeloid Leukemia. *J Clin Oncol* 2003; **21**: 4642–4649.
19 van Dongen JJ, Macintyre EA, Gabert JA, Delabesse E, Rossi V, Saglio G *et al.* Standardized RT-PCR analysis of fusion gene transcripts from chromosome aberrations in acute leukemia for detection of minimal residual disease. Report of the BIOMED-1 Concerted Action: investigation of minimal residual disease in acute leukemia. *Leukemia* 1999; **13**: 1901–1928.
20 Gabert J, Beillard E, van der Velden VH, Bi W, Grimwade D, Pallisgaard N *et al.* Standardization and quality control studies of 'real-time' quantitative reverse transcriptase polymerase chain reaction of fusion gene transcripts for residual disease detection in leukemia - a Europe Against Cancer program. *Leukemia* 2003; **17**: 2318–2357.
21 Kaplan EL, Meier P. Non-parametric estimation from incomplete observation. *J Am Stat Assoc* 1958; **53**: 457–481.
22 Cooley TA, Leisenring W, Crowley J, Storer BE. Estimation of failure. Probabilities in the presence of competing risks: new representation of old estimators. *Stat Med* 1999; **18**: 695–706.
23 Gray R. A class of K-sample tests for comparing the cumulative incidence of a competing risk. *Ann Stat* 1988; **16**: 1141–1151.
24 Cox D. Regression models and life tables. *J R Stat Soc B* 1972; **34**: 187–220.
25 Pacilli L, Lo Coco F, Ramadan SM, Gianni L, Pingi A, Remotti D *et al.* Promyelocytic lymphoma of the spine: a case report and review of the literature. *Adv Hematol* 2010; **2010**: 137608.
26 Lou Y, Suo S, Tong Y, Tong H, Qian W, Meng H *et al.* Outcomes and prognostic factors of first relapsed acute promyelocytic leukemia patients undergoing salvage therapy with intravenous arsenic trioxide and chemotherapy. *Ann Hematol* 2014; **93**: 941–948.
27 Au WY, Tam S, Fong BM, Kwong YL. Elemental arsenic entered the cerebrospinal fluid during oral arsenic trioxide treatment of meningeal relapse of acute promyelocytic leukemia. *Blood* 2006; **107**: 3012–3013.
28 De Botton S, Sanz MA, Chevret S, Dombret H, Martin G, Thomas X *et al.* Extramedullary relapse in acute promyelocytic leukemia treated with all-trans retinoic acid and chemotherapy. *Leukemia* 2006; **20**: 5–41.
29 Montesinos P, Díaz-Mediavilla J, Debén G, Prates V, Tormo M, Rubio V *et al.* Central nervous system involvement at first relapse in patients with acute promyeolcytic leukemia treated with all-trans retinoic acid and anthracycline monochemotherapy without intrathecal prophylaxis. *Haematologica* 2009; **94**: 1242–1249.
30 Wang H, Cao F, Li J, Li L, Li Y, Shi C *et al.* Arsenic trioxide and mannitol for the treatment of acute promyelocytioc leukemia relapse in central nervous system. *Blood* 2014; **124**: 1998–2000.
31 Douer D, Hu W, Giralt S, Lill M, DiPersio J. Arsenic trioxide (trisenox) therapy for acute promyelocytic leukemia in the setting of hematopoietic stem cell transplantation. *Oncologist* 2003; **8**: 132–140.
32 De Botton S, Fawaz A, Chevret S, Dombret H, Thomas X, Sanz M *et al.* Autologous and allogeneic stem-cell transplantation as salvage treatment of acute promyelocytic leukemia initially treated with all-trans-retinoic acid: a retrospective analysis of the European acute promyelocytic leukemia group. *J Clin Oncol* 2005; **23**: 120–126.
33 Thirugnanam R, George B, Chendamarai E, Lakshmi KM, Balasubramanian P, Viswabandya A *et al.* Comparison of clinical outcomes of patients with relapsed acute promyelocytic leukemia induced with arsenic trioxide and consolidated with either an autologous stem cell transplant or an arsenic trioxide-based regimen. *Biol Blood Marrow Transplant* 2009; **15**: 1479–1484.
34 Yanada M, Tsuzuki M, Fujita H, Fujimaki K, Fujisawa S, Sunami K *et al.* Phase 2 study of arsenic trioxide followed by autologous hematopoietic cell transplantation for relapsed acute promyelocytic leukemia. *Blood* 2013; **121**: 3095–3102.
35 Pemmaraju N, Tanaka MF, Ravandi F, Lin H, Baladandayuthapani V, Rondon G *et al.* Outcomes in patients with relapsed or refractory acute promyelocytic leukemia treated with or without autologous or allogeneic hematopoietic stem cell transplantation. *Clin Lymphoma Myeloma Leuk* 2013; **13**: 485–492.
36 Fujita H, Asou N, Iwanaga M, Hyo R, Nomura S, Kiyoi H *et al.* Role of hematopoietic stem cell transplantation for relapsed acute promyelocytic leukemia: a retrospective analysis of JALSG-APL97. *Cancer Sci* 2013; **104**: 1339–1345.
37 Holter Chakrabarty JL, Rubinger M, Le-Rademacher J, Wang HL, Grigg A, Selby GB *et al.* Autologous is superior to allogeneic hematopoietic cell transplantation for acute promyelocytic leukemia in second complete remission. *Biol Blood Marrow Transplant* 2014; **20**: 1021–1025.
38 Kohno A, Morishita Y, Iida H, Yanada M, Uchida T, Hamaguchi M *et al.* Hematopoietic stem cell transplantation for acute promyelocytic leukemia in second or third complete remission: a retrospective analysis in the Nagoya Blood and Marrow Transplantation Group. *Int J Hematol* 2008; **87**: 210–216.
39 Lo-Coco F, Avvisati G, Vignetti M, Thiede C, Orlando SM, Iacobelli S *et al.* Retinoic acid and arsenic trioxide for acute promyelocytic leukemia. *N Engl J Med* 2013; **369**: 111–121.
40 Fenaux P, Chevret S, Guerci A, Fegueux N, Dombret H, Thomas X *et al.* Long-term follow-up confirms the benefit of all-trans retinoic acid in acute promyelocytic leukemia. European APL group. *Leukemia* 2000; **14**: 1371–1377.