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Conflict of interest disclosure

The authors declare no competing financial interests.

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Supporting Information

Additional supporting information may be found online in the Supporting Information section at the end of the article.

Figure S1. Analysis of $CD4^+$ and $CD8^+$ T lymphocyte V-beta repertoire in BM and PB of healthy donors as compared with MDS patients.

Data S1. Methods and Materials.

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Arsenic trioxide and all-trans retinoic acid treatment for childhood acute promyelocytic leukaemia

Acute promyelocytic leukaemia (APL) has evolved from being frequently fatal to a highly curable leukaemia through the combination of all-trans retinoic acid (ATRA) and anthracycline-based chemotherapy, with remission and event-free survival (EFS) rates in children approaching 95% and 80%, respectively (Testi *et al*, 2005; Imaizumi *et al*, 2011).

Recent experience in adults with standard-risk (SR) APL [i.e., white blood cell (WBC) count $<10 \times 10^9$/l at diagnosis] showed that combining arsenic trioxide (ATO) with ATRA results in impressive cure rates, with a milder toxicity profile when compared to conventional chemotherapy (Lo-Coco *et al*, 2013; Burnett *et al*, 2015; Platzbecker *et al*, 2017).

We analysed 18 unselected, consecutive cases of newly diagnosed paediatric APL treated using the ATO-ATRA combination for both induction and consolidation therapy in 7 Italian centres from October 2014 to July 2017. Diagnosis of APL was established in all patients by detection of the *PML-RARA* fusion and/or demonstration of the t(15;17) translocation. Patients were defined as SR if the WBC count at diagnosis was $<10 \times 10^9$/l, or high-risk (HR) when WBC count was $\geq 10 \times 10^9$/l, according to the modified Sanz criteria (Sanz *et al*, 2000).

Treatment for both SR and HR patients was based on the protocol scheme described for SR adult patients (Lo-Coco *et al*, 2013). Details on therapy administered and on definitions used are reported in the Data S1.

Baseline patient characteristics are summarized in Table I; 16/18 patients were SR group, while the remaining 2 children had HR disease. The observed toxicities/adverse events are summarized in Table II. No fatal events occurred; haematological toxicity was observed during induction therapy in 15 patients (83·3%), with grade 3–4 neutropenia occurring in 12 patients (66·6%) and grade 3–4 thrombocytopenia in 10 patients (55·5%). No further haematological toxicity was observed during consolidation courses. Hepatotoxicity occurred in 9 patients (50%) and was limited to a rise in serum transaminase levels, with grade 3 toxicity being recorded in one case only. Three patients experienced prolongation of the QTc interval, which was successfully managed with temporary ATO discontinuation.

Eleven of 18 patients (9 SR and 2 HR patients) (61·1%) developed sustained hyperleucocytosis during induction, requiring administration of hydroxycarbamide. The median WBC peak was 51×10^9/l (range 28–113), recorded at a median time of 10 days (range 5–19) after starting induction therapy. Four additional SR patients experienced milder leucocytosis (WBC count 10–20 $\times 10^9$/l), which resolved spontaneously. One HR patient presented on day 10 with interstitial pulmonary infiltrates, mild weight gain and musculoskeletal pain. Dexamethasone was started at onset of signs/symptoms, with rapid improvement and complete resolution of the clinical picture.

Pseudotumor cerebri was observed in 2 patients during consolidation therapy and resolved after temporary ATRA discontinuation. Median hospital-stay during induction therapy was 31·5 days (range 9–43), while consolidation courses were administered on an outpatient basis.

All patients completed the treatment protocol and achieved haematological complete remission (CR) at the end of induction. The median time to haematological CR was 38·5 days (range, 28–53).

All patients achieved molecular CR after the third ATO consolidation course. With a median follow-up of 24 months (range 9–42), all patients are alive and in molecular CR.

Table I. Patient characteristics at diagnosis.

	N or median	(% or range)
Age at diagnosis (years)	13·9	(4·8–17·5)
Gender		
Males	9	(50%)
Females	9	(50%)
Risk group		
SR	16	(89%)
HR	2	(11%)
WBC count at diagnosis ($\times 10^9$/l)	4·53	(0·86–33·7)
SR	4·51	(0·86–9·4)
HR	11·7 and 33·7	
Platelet count at diagnosis ($\times 10^9$/l)	34	(8–170)
SR	29	(8–170)
HR	39 and 58	
Coagulopathy		
Yes	13	(72%)
No	5	(28%)
PML-RARA isoform		
Bcr1	8	(44·4%)
Bcr2	2	(11·1%)
Bcr3	8	(44·4%)
FLT3-ITD mutation		
Yes	3	(17%)
No	12	(66%)
Missing data	3	(17%)

HR, high-risk; ITD, internal tandem duplication; N, number; SR, standard-risk; WBC, white blood cell.

Table II. Toxicities/adverse events observed in our population.

	Grade 1–2 *N* (%)	Grade 3–4 *N* (%)	Total *N* (%)
Haematological toxicity			
Neutropenia	0 (0)	12 (66·6)	12 (66·6)
Thrombocytopenia	3 (16·6)	10 (55·5)	13 (72·2)
Anaemia	4 (22·2)	9 (50)	13 (72·2)
Extra-haematological toxicity			
Hepatic toxicity	8 (44·4)	1 (5·5)	9 (50)
QTc prolongation	3 (16·6)	0 (0)	3 (16·6)
			15 (83·3)
Hyperleucocytosis during induction			
WBC count $\geq 20 \times 10^9$/l			11 (61·1)
WBC count 10–20 $\times 10^9$/l			4 (22·2)
			1 (5·5)
Differentiation syndrome (suspected*)			
Pseudotumor cerebri			2 (11·1)

Adverse events were graded according to the Common Terminology Criteria for Adverse Events (CTCAE) version 4.03 (https://www.eortc.be/services/doc/ctc/CTCAE_4.03_2010-06-14_QuickReference_5x7.pdf). N, number of patients; WBC, white blood cell.

*Clinical picture characterized by interstitial pulmonary infiltrates, mild weight gain, peripheral oedema, and musculoskeletal pain, without fever, respiratory distress, pleural/pericardial effusion or hypotension.

Despite the dramatic improvement achieved in frontline therapy of childhood APL with the combination of ATRA and anthracycline-based regimens, relapse and chemotherapy-associated side effects still represent a non-negligible cause of treatment failure and morbidity. The remarkable results obtained in the randomized clinical trial APL0406 in adults with SR APL showed a clear advantage of ATO-ATRA over ATRA-chemotherapy in terms of overall survival, EFS, cumulative incidence of relapse and haematological toxicity, supporting the use of the ATO-ATRA combination as the new standard of care for newly diagnosed SR adults (Lo-Coco *et al*, 2013; Platzbecker *et al*, 2017). A subsequent randomized study conducted by the United Kingdom National Cancer Research Institute (NCRI) confirmed the advantage, in terms of EFS and relapse rate, of the ATO-ATRA approach over ATRA-chemotherapy, suggesting its feasibility also in HR patients, who were given one dose of the CD33-targeted immunoconjugate gemtuzumab ozogamicin (GO) in the first days of induction (Burnett *et al*, 2015).

A frontline treatment approach based on the ATO-ATRA combination has been explored in a limited number of paediatric patients (Cheng *et al*, 2013; Creutzig *et al*, 2017). A retrospective case series including 11 children with SR APL given a frontline ATO-ATRA protocol was recently reported by the AML-Berlin-Frankfurt-Münster group (Creutzig *et al*, 2017). All patients achieved molecular CR after a median time of 10 weeks (range 7–20). Differences from the APL0406 protocol included a 1-week break of ATRA after the first 14 days of treatment and a delayed start of ATO (given on day 10) to avoid the risk of hyperleucocytosis. Nonetheless, all patients experienced hyperleucocytosis, which required the administration of low-dose cytarabine in 3 cases (*plus* liposomal daunorubicin in one case). The recently published COG-AAML0631 protocol for newly diagnosed APL, including 101 children given induction therapy with Idarubicin and ATRA, combined ATO and ATRA for consolidation-1 in SR and HR patients (Kutny *et al*, 2017). This trial demonstrated that ATO consolidation allowed significant reduction in anthracycline doses, while maintaining excellent results, with a 3-year EFS of 91%.

Based on the excellent results obtained in adults, we tested in 7 Italian centres the chemotherapy-free approach for children with newly diagnosed APL. The most common adverse effects were either transient/self-limiting or resolved with temporary therapy discontinuation. Notably, all 18 patients, including the 2 children with HR APL, achieved both haematological and molecular CR without any chemotherapy other than hydroxycarbamide. Although 2 HR children only were included in our series, their favourable outcome suggests that this subset of patients might also benefit from a chemotherapy-free approach.

To the best of our knowledge, this is the largest European series of children with APL treated with a frontline chemotherapy-free approach for both induction and consolidation therapy. An open label, prospective multicentre European trial from the International Consortium for Childhood APL (ICC APL Study 02) will be started soon to validate these results in children with newly diagnosed APL.

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Authorship Contributions

F.L. and F.L.C. designed the treatment schemes; L.S. and F.L. wrote and edited the manuscript; L.S. and C.G. collected and analysed the data; F.L.C. and D.D. performed *PML-RAR*α analysis for molecular diagnosis and monitoring; F.L.C. performed *FLT3-ITD* molecular analysis, and contributed to critical review of the manuscript; L.S., F.L., N.S., M.C.P., C.M., M.Z., R.C., K.G., and A.M.T., were involved in the clinical management of patients and data collection. All authors contributed to the intellectual content of this paper and approved the final manuscript.

Disclosure competing interests statement

The Authors declare no competing financial interest related to this work.

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Supporting Information

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Data S1. Treatment details and definitions.

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Use of PD-1 (PDCD1) inhibitors for the treatment of Richter syndrome: experience at a single academic centre

An aggressive lymphoma in a patient with chronic lymphocytic leukaemia (CLL), known as Richter syndrome (RS), has a poor prognosis and is an important unmet clinical need (Richter, 1928; Parikh *et al*, 2014). Standard treatment with chemoimmunotherapy is unsuitable for many patients who are unfit for these intensive regimens or have disease features predicting poor response, such as complex CLL karyotype or relapse after prior chemoimmunotherapy, leaving no good options for these patients. (Tsimberidou *et al*, 2006; Rossi *et al*, 2011; Parikh *et al*, 2014; Rogers *et al*, 2018).

Recently, the programmed death receptor-1 (PD-1, also termed PDCD1) blocking antibodies, pembrolizumab and nivolumab, have shown efficacy in prospective clinical trials