ATO/ATRA/Anthracycline-Chemotherapy Sequential Consolidation Achieves Long-Term Efficacy in Primary Acute Promyelocytic Leukemia

Zi-Jie Long⁰¹⁳, Yuan Hu⁰¹⁹, Xu-Dong Li¹, Yi He¹, Ruo-Zhi Xiao¹, Zhi-Gang Fang¹, Dong-Ning Wang¹, Jia-Jun Liu¹, Jin-Song Yan², Ren-Wei Huang¹*, Dong-Jun Lin¹*, Quentin Liu¹,²*

¹ Department of Hematology, Third Affiliated Hospital, Sun Yat-sen University, Sun Yat-sen Institute of Hematology, Sun Yat-sen University, Guangzhou, China, ² Institute of Cancer Stem Cell, Dalian Medical University, Dalian, China, ³ Department of Hematology, Second Affiliated Hospital, Dalian Medical University, Dalian, China

Abstract

The combination of all-trans retinoic acid (ATRA) and arsenic trioxide (As₂O₃, ATO) has been effective in obtaining high clinical complete remission (CR) rates in acute promyelocytic leukemia (APL), but the long-term efficacy and safety among newly diagnosed APL patients are unclear. In this retrospective study, 45 newly diagnosed APL patients received ATRA/chemotherapy combination regimen to induce remission. Among them, 43 patients (95.6%) achieved complete remission (CR) after induction therapy, followed by ATO/ATRA/anthracycline-based chemotherapy sequential consolidation treatment with a median follow-up of 55 months. In these patients, the estimated overall survival (OS) and the relapse-free survival (RFS) were 94.4%±3.9% and 94.6±3.7%, respectively. The toxicity profile was mild and reversible. No secondary carcinoma was observed. These results demonstrated the high efficacy and minimal toxicity of ATO/ATRA/anthracycline-based chemotherapy sequential consolidation treatment for newly diagnosed APL in long-term follow-up, suggesting a potential frontline therapy for APL.

Introduction

Acute promyelocytic leukemia (APL), characterized by the t(15, 17) chromosomal translocation and leukemogenic PML-RARα fusion protein, is accumulated of abnormal promyelocytes in the bone marrow and causes severe bleeding tendency [1]. The treatment of APL with chemotherapy achieved complete remission (CR) in two-thirds of newly diagnosed patients, however, the 5-year disease-free survival (DFS) was still very poor [1–3]. The induction of all-trans retinoic acid (ATRA) in the treatment and optimization of the anthracycline-based regimens resulted in terminal differentiation of APL cells with a 90–95% CR and the 5-year DFS up to 74% [1,4,5], although approximately 5–30% of patients developed disease recurrence [6].

As one of the most potential drugs in APL, arsenic trioxide (As₂O₃, ATO) targets PML/RARα and exerts dose-dependent dual effects on APL cells, with low concentrations inducing differentiation and high concentrations triggering apoptosis [7]. Since 1990s, the use of ATO has improved the clinical benefit of refractory or relapsed as well as newly diagnosed APL [8–11]. ATO injection for APL patients who developed disease recurrence or failed to respond to standard treatment was later approved by the US FDA. Moreover, molecular remission is obtainable in patients from 72% to 91% after CR by ATO alone [12,13]. Strong synergistic anti-leukemic effects of ATO in combination with ATRA were found in both APL cell lines and APL animal models, with induction catabolism of the PML-RARα fusion protein [14–17]. Importantly, previous clinical trials showed that the combination of ATO and ATRA yielded a longer survival rate compared to either ATRA or ATO monotherapy [18–23]. Moreover, ATO consolidation therapy spared anthracycline exposure [24], and improved both event-free survival (EFS) and overall survival (OS) in newly diagnosed APL [25]. Yet, a standard ATO/ATRA consolidation regimen for newly diagnosed APL remains to be further validated.

In this retrospective study, ATRA/chemotherapy combination regimen was applied to induce remission for newly diagnosed APL patients. A regimen consisting of ATO, ATRA and anthracycline-based chemotherapy was used sequentially as consolidation therapy for the patients who obtained CR. The long-term efficacy and safety of ATO/ATRA/anthracycline-based chemotherapy consolidation regimen were evaluated.

Methods

Patients

This retrospective study consisted of 45 patients with newly diagnosed APL in the Third Affiliated Hospital, Sun Yat-sen University, from March 1, 2000 to August 31, 2012. The median age was 29 years (10–62 years). Pertinent patient clinical reports of this study were obtained with patients’ written consent and the approval of the Ethical Board of The Third Affiliated Hospital,
Sun Yat-sen University ([2013]2-69). Parental written consent was obtained for underage participants.

APL diagnosis was established according to clinical presentations, morphological criteria of the French-American-British classification, cytogenetic assay for t (15; 17) (q22; q21), and RT-PCR analysis for PML-RARα transcripts. The exclusion criteria for this retrospective study included: dysfunction of liver or kidney; any heart diseases or cardiac functional insufficiency; patients who died before initiation of the therapy. Standard induction therapy was administered for the 45 newly diagnosed APL patients (Figure 1). Two patients died during induction treatment. The remaining 43 patients received consolidation therapy. The clinical features of patients were described in Table 1.

Remission Induction Therapy

Induction therapy for these newly diagnosed patients with APL was a combination of ATRA and daunorubicin plus cytarabine. Once the diagnosis was suspected on the basis of clinical features and the peripheral blood smear, ATRA was administered orally at 40 mg/m²/day (divided into two equal doses) until CR was achieved. Patients with WBC counts ≥10×10⁹/L additionally received hydroxycarbamide orally until the WBC count was down to less than 10×10⁹/L. ATRA was continued for 3 to 15 days to ameliorate the coagulopathy before initiating chemotherapy (daunorubicin 40 mg/m²/day for 3 days, cytarabine 100 mg/q12h for 7 days).

![Figure 1. A chart review of patients treated with standard of induction and consolidation therapy.](doi:10.1371/journal.pone.0104610.g001)
Supportive Care

During induction of remission, examinations including whole peripheral blood cell counts, renal and hepatic function tests were performed. Coagulation and fibrinolysis parameters including fibrinogen, D-dimers, fibrin degradation product (FDP), prothrombin time (PT), and activated partial thromboplastin time (APTT) were monitored to identify the requirement of platelet, fresh plasma, or cryoprecipitate transfusions. Supportive treatment was based on maintaining platelet counts \( \geq 30 \times 10^9/L \) until coagulopathy disappearance. Electrocardiogram and sonography were used for monitoring the cardiac function for patients. APL differentiation syndrome (APLDS) was treated with prednisone or dexamethasone until clear resolution of symptoms. Drug toxicities were documented using the National Cancer Institute-Common Toxicity Criteria, version 3.0. Symptomatic therapy was performed for the side effects of ATO, ATRA and anthracycline.

Consolidation Therapy

Patients were monitored to confirm that the bone marrow morphology and recovery of peripheral blood cell counts. Consolidation therapy included 6 courses was initiated once CR was achieved, and each course included three consecutive regimens: (1) ATO, 10 mg/day for 14 days intravenously; (2) ATRA, 25 mg/m^2/day for 30 days orally; (3) anthracycline-based regimens: daunorubicin (40 mg/m^2/day), or idarubicin (8 mg/m^2/day), or pirarubicin (25 mg/m^2/day) for 3 days plus cytarabine 100 mg/q12 h for 5 days. The three regimens of consolidation therapy were administered sequentially every month in the first year after achieving CR. In the second year, each regimen of consolidation therapy was administered sequentially every two months. Six courses were given totally.

All patients received intrathecal therapy (methotrexate 15 mg, cytarabine 50 mg, dexamethasone 8 mg) when CR was achieved. Prophylaxis was performed 4–6 times altogether.

Response Definition

CR was defined according to clinical presentations and morphological criteria, including cellular bone marrow blasts and abnormal promyelocytes \( \leq 5\% \) with an absolute neutrophil count \( \geq 1.0 \times 10^9/L \) and platelet count \( \geq 100 \times 10^9/L \). Clinical recurrence was defined as the presence of \( \geq 5\% \) blasts, or abnormal promyelocytes in the bone marrow, or the appearance of leukemic cells in peripheral blood, or abnormal promyelocytes in cerebrospinal fluid (CSF). RT-PCR for the PML-RAR\(_a\) fusion transcript was performed on the bone marrow follow-up every 2 months for monitoring molecular remission. After molecular remission, the examination was still performed every 3 months for monitoring relapse.

Patients with chronic hepatitis B were treated with lamivudine or telbivudine for prevention of virus activation.

Table 1. Clinical data of the patients.

|                      | N = 45 |
|----------------------|--------|
| Gender, male/female  | 20/25  |
| Median age, years    | 29 (10–62) |
| WBC, \( \times 10^9/L \) | 2.3 (0.2–47.5) |
| Median Hb, g/L       | 81.0 (38.0–120.0) |
| Median platelet, \( \times 10^9/L \) | 23.0 (5.0–120.0) |
| Clinical CR          | 95.6%  |
| Median days to clinical CR | 30 (20–60) |
| Median months to molecular CR | 6 (2–12) |

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Figure 2. Survival analysis. The OS for all 45 patients.
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Statistical Analysis

OS was defined as the time from the initiation of induction therapy to death. RFS was defined as the time from CR to relapse. Survival analysis was performed using Kaplan-Meier estimate methods. Statistical analysis was performed using SPSS16.0 for windows software.

Results

Outcomes

As seen in Table 1, among total 45 patients, 43 (95.6%) achieved CR in remission introduction therapy. The median time to achieve CR was 30 days (range: 20–60 days). Two patients suffered from early death within 15 days during the induction
therapy due to intracranial hemorrhage (1 case), or acute tumor lysis syndrome (1 case). For the 43 patients who entered CR, all received ATO/ATRA/anthracycline-based chemotherapy for consolidation therapy. The median follow-up was 55 months (range: 6–150 months), and the median months to molecular CR was 6 months (range: 2–12 months). Till the end of this study, 41 patients remained in good clinical and molecular remission. Two patients relapsed: one presented with central nervous system (CNS) leukemia in the 27th month and the other developed full APLDS was diagnosed in 2 patients (4.7%). Other side effects of ATRA, such as skin reactions (19 patients, 44.2%), headache (13 patients, 30.2%), gastrointestinal tract reactions (6 patients, 14.0%) and fever (4 patients, 9.3%), were mild and overcame by administration of symptomatic medication. During consolidation, 6 of 41 patients developed tolerable and reversible grade I liver dysfunction and 1 patient developed grade II liver dysfunction, whereas no grade III–IV liver toxicity was observed. Hepatic function returned to normal in all of these patients after supportive therapy. No one needed termination of ATO therapy because of severe liver damage. Therapy-related neutropenia were observed in 8 patients (18.6%). One 62-year-old patient presented with chronic cardiac insufficiency in the 18th month after CR, which might be due to the accumulation of anthracycline for the elderly. In addition, all the 8 hepatitis B patients did not show any virus reactivation during consolidation.

**Table 2. Toxicity profile.**

| Toxicity          | N = 43 | Grade I | Grade II | Grade III | Grade IV | Skin reaction | Headache | Neutropenia | Gastrointestinal reaction | Cardiac arrhythmia | APLDS | Fever |
|-------------------|--------|---------|----------|-----------|----------|---------------|----------|-------------|--------------------------|-------------------|-------|-------|
| Hepatotoxicity    | 7 (16.3%) | 6 (14.0%) | 1 (2.3%) | 0 (0%)    | 0 (0%)    | 19 (44.2%)    | 13 (30.2%)| 8 (18.6%)   | 6 (14.0%)               | 1 (2.3%)          | 2 (4.7%)| 4 (9.3%)|
| Grade II          | 6 (14.0%) | 1 (2.3%) |          |           |           |               |          |             |                          |                   |       |       |
| Grade III         | 1 (2.3%)  |          |          |           |           |               |          |             |                          |                   |       |       |
| Grade IV          | 0 (0%)    |          |          |           |           |               |          |             |                          |                   |       |       |
| Skin reaction     | 19 (44.2%)|          |          |           |           |               |          |             |                          |                   |       |       |
| Headache          | 13 (30.2%)|          |          |           |           |               |          |             |                          |                   |       |       |
| Neutropenia       | 8 (18.6%) |          |          |           |           |               |          |             |                          |                   |       |       |
| Gastrointestinal reaction | 6 (14.0%) |          |          |           |           |               |          |             |                          |                   |       |       |
| Cardiac arrhythmia| 1 (2.3%)  |          |          |           |           |               |          |             |                          |                   |       |       |
| APLDS             | 2 (4.7%) |          |          |           |           |               |          |             |                          |                   |       |       |
| Fever             | 4 (9.3%) |          |          |           |           |               |          |             |                          |                   |       |       |

**Discussion**

ATRA in combination with anthracycline-based chemotherapy is considered as the standard for the induction and consolidation therapy of newly diagnosed APL. However, cumulative incidence of relapse still occurs in one third of the patients who have obtained CR. ATO induced catabolism of the PML-RARα fusion protein, demonstrating an effective targeted therapy in APL. In 1990s, the possibility of using a triad of chemotherapy, ATRA, and ATO for newly diagnosed patients in APL was discussed at a meeting in Shanghai. Then studies in the mouse model showed that this combination could dramatically prolong the survival or even eradicate disease. These results encouraged physicians to conduct new therapeutic approaches based on ATO/ATRA/anthracycline-based chemotherapy combination for the treatment of newly diagnosed APL patients.

Indeed, since the introduction of ATRA/ATO-based combination treatment for newly diagnosed APL and recurrence, the CR rate and the 5-year DFS have been greatly improved [18–23,26]. In this study, the ATRA/chemotherapy combination regimen was administered to induce remission, and the ATO plus ATRA and anthracycline-based chemotherapy consolidation regimen was used to maintain long-term efficacy for newly diagnosed APL patients. In 45 de novo patients, CR was achieved in 43 patients (95.6%), whereas the median time to achieved CR was 30 days. The estimated 3-year OS rate for all patients was 90.2%±4.7%. For patients who achieved CR (n = 43), the OS and RFS rates were 94.4%±3.9% and 94.6±3.7%, respectively. Our data were consistent with recent studies [23], which reported a long-term outcome in the ATRA/ATO-based regimen.

The therapeutic benefit of ATO as a single agent for the treatment of APL has been reported previously [27,28], thus using ATO as the post-remission therapy for the APL patients in CR was reasonable. Importantly, ATO consolidation produced a good survival rate no matter which method was used in CR induction and eliminated the need for maintenance therapy [29–31]. However, the relatively high incidence of ATO-induced hepatotoxicity during remission induction remains unclear and worthy of note, though the side effects of ATO were considered to be moderate. Reversible grade III–IV hepatotoxicity was seen in a small proportion of patients [27]. Overtreatment in the majority of patients was potentially associated with a risk of treatment-related death during early disease remission as well as longer-term risks of secondary carcinoma or anthracycline-related cardiomyopathy. Thus in the present study, ATRA-based induction regimen was applied and ATO was not added to the remission regimen. Either the daily or the total dosage of ATO for consolidation was minimal (10 mg/day for 14 days each course), which APL patients could benefit from ATO by consolidation without overtreatment during each course. In fact, during the consolidation, no grade III–IV hepatotoxicity was documented in our patients. Only 7 patients developed tolerable and reversible grade I–II liver dysfunction, and their hepatic function returned to normal after consolidation therapy. Other side effects were minimal during post-remission treatment. Another major concern associated with long-term exposure to ATO is secondary tumors, and we found no cases in the present study developed secondary tumors. Besides, our analysis showed that incorporation of ATRA drastically achieved long-term efficacy. Importantly, patients in our study showed a very low incidence of APLDS (4.7%). While the dosage of ATO was relatively small, APL patients could benefit from the consolidation with ATO and ATRA, thus usage of ATO/ATRA combination as the post-remission therapy for the APL patients in CR contributed to high efficacy and low side effects.

The therapeutic benefit of ATRA/ATO use in relapsed APL has been described previously [32–35]. However, in a randomized study of 10 cases, the ATRA/ATO combination regimen failed to induce synergistic effect [36]. In our study, the beneficial effects...
| Clinical Studies | No. of patients | Age (median) | Sanz Risk (low/ int/high) | Induction Therapy | CR | Consolidation Therapy | Maintenance Therapy | Survival Outcome |
|------------------|----------------|-------------|---------------------------|-------------------|----|----------------------|---------------------|------------------|
| Long ZJ, et al.  | 45 (20/25)     | 29 (10–62)  | low/ 38; high 7           | ATRA+DNR+Ara-C    | 95.6% | ATO+ATRA+IDA+Ara-C, 6 courses | 3-year OS 90.2%, RFS 94.6% |
| Zhang YM, et al. | 33 (18/15)     | 65 (60–79)  | 6/22/5                    | ATO               | 87.9% | ATO, 4 years          | 10-year OS 69.3%, DFS 64.8%, CSS 84.8% |
| Lo-Coco F, et al.| A: 77 (40/37); B: 79 (56/43) | A: 44.6 (19.1–70.2); B: 46.6 (18.7–70.2) | A: low/int 3/4/4; B: low/int 27/52 | A: ATRA+ATO; B: ATRA+IDA | A: 100%; B: 95% | A: ATO+ATRA, 28 weeks; B: ATRA+NOT/MTZ, 3 cycles | 2-year OS 99%, DFS 97%; B: 2-year OS 91%, DFS 90% |
| Iland HU, et al. (ARMLA) | 124 (62/62) | 44 (3–78) | 32/67/24 | ATRA+IDA+ATO | 95% | ATO+ATRA, 2 cycles | ATRA, MTX, 6-MP, 8 cycles |
| Avvisati G, et al. 2011 (AIDA 0493) | 828 (438/390) | 37.2 (1.4–74.7) | 157/432/231 | ATRA+IDA | 94.3% | IDA+Ara-C, MTZ+VP-16, IDA+Ara-C+6-TG, 3 courses | 6-MP, MTX, ATRA, 2 years |
| Sanz MA, et al. 2010 (LPA2005) | 402 (209/193) | 42 (3–83) | 84/200/118 | ATRA+IDA | 99%/95%/83% | IDA, ATRA, MTZ, Ara-C, 3 courses | 4-year DFS 90% (93%/92%/ 82%), OS 88% (96%/91%/ 79%) |
| Powell BL, et al. 2010 (C9710) | A: 244 (123/121); B: 237 (124/113) | 15–60 year 207/197; >60 year 37/40 | A: 69/120/55; 6/7/112/58 | A: ATRA+ARA-C+DNR; B: ATRA+ARA-C+DNR | A: 90%; B: 90% | A: ATO, 2 cycles (+ATRA+DNR), 2 cycles; B: (+ATRA+DNR), 2 cycles | ATRA+6-MP/MTX, 1 year |
| Lengfelder E, et al. 2009 142 (59/83) (AML) | 209/113 | 207/197; >60 year 37/40 | ATRA+ATO | 94.1% | DNAR+Ara-C, Ara-C pulse, HHT+Ara-C, 3 cycles | ATRA, ATO, MTX/6-MP, 5 cycles |
| Asou N et al. 2007 (APL97) | 283 (158/125) | 48 (15–70) | low/int 232; high 51 | ATRA+IDA/Ara-C | 94% | MTZ+Ara-C, DNR+VP-16+Ara-C, IDA+Ara-C, 3 courses | 6-year DFS 68.5%, OS 83.9% |

Abbreviations: low/int/high: low/intermediate/high; OS: overall survival; DFS: disease-free survival; CSS: cause-specific survival; FFS: failure-free survival; Ara-C: cytarabine; BHAC: behenoyl Ara-C; DNR: daunorubicin; IDA: idarubicin; MTX: methotrexate; 6-MP: mercaptopurine; MTZ: mitoxantrone; VP-16: etoposide; VDS: vindesin; ACR: aclacinomycin; 6-TG: 6-thioguanine; HHT: homoharringtonine; CTX: cyclophosphamide.
were observed in the newly diagnosed APL, in contrast to that report. The reason might be that majority of the relapsed patients lost sensitivity to ATRA due to previous exposure, making it difficult to expect a full efficacy of the synergism between ATRA and ATO in those patients. In addition, parts of recent studies about different models of patients were summarized in Table 3 [23,25,31,37–42] to make a comparison and we found that there was no strong evidence about the recommended strategy for different risk groups. However, the addition of ATO was proved to improve the long-term survival of patients with different risks, which gave support to our present study.

Mechanically, ATRA and ATO targets PML/RARa and exerts dose-dependent differentiation and apoptosis. Microarray, proteomics, and bioinformatics revealed that synergistic effect in combination therapy was due to transcriptional remodeling induced by ATRA-induced differentiation and ATO-related proteome level change. Importantly, enhanced degradation of PML-RARa might be considered for the efficacy of combination therapy in patients: ATO targeted PML, while ATRA aimed to RARa. Besides RA signaling and ubiquitin-proteasome pathway, some self-renewal and differentiation related molecules were newly revealed to be involved in the ATO/ATRA synergistic effect, such as c-myc, Bmi-1 [14,43]. Thus, further studies should attempt to identify the network by which ATO/ATRA regulates in APL cells.

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Author Contributions
Conceived and designed the experiments: QL RWJ DJL. Performed the experiments: XDL YH RZ JG DNW JJL. Analyzed the data: ZJL YH XDL. Contributed reagents/materials/analysis tools: QL DJL. Wrote the paper: ZJL YH JSY.

References
1. Wang ZY, Chen Z (2008) Acute promyelocytic leukemia: From highly fatal to highly curable. Blood 111: 2500–2513.
2. Cunningham I, Gee TS, Reich LM, Kempin SJ, Naval AN, et al. (1989) Acute promyelocytic leukemia: treatment results during a decade at Memorial Hospital. Blood 73: 1116–1122.
3. Sanz MA, Jarque I, Martin G, Lorenzo I, Martinez J, et al. (1988) Acute promyelocytic leukemia: therapy results and prognostic factors. Cancer 61: 7–13.
4. Huang ME, Ye YC, Chen SR, Chai JR, Lu JX, et al. (1988) Use of all-trans retinoic acid in the treatment of acute promyelocytic leukemia. Blood 72: 567–572.
5. Tallman MS, Anderssen JW, Schaffer CA, Appelbaum FR, Feusner JH, et al. (2002) All-trans retinoic acid in acute promyelocytic leukemia: Long-term outcome and prognostic factor analysis from the North American Intergroup protocol. Blood 100: 4298–4302.
6. Tallman MS (2007) Treatment of relapsed or refractory acute promyelocytic leukemia. Best Pract Res Clin Haematol 20: 57–63.
7. Chen GQ, Shi XG, Tang W, Xiong SM, Zhu J, et al. (1997) Use of arsenic trioxide in the treatment of acute promyelocytic leukemia (APL): As2O3 exerts dosedependent dual effects on APL cells. Blood 89: 3354–3355.
8. Shen ZX, Chen GQ, Ni JH, Li XS, Xiong SM, et al. (1997) Use of arsenic trioxide (As2O3) in the treatment of acute promyelocytic leukemia (APL): II. Clinical efficacy and pharmacokinetics in relapsed patients. Blood 90: 3354–3360.
9. Sun HD, Ma L, Hu XC, Zhang TD (1992) Ai-Lin I treated 32 cases of acute promyelocytic leukemia. Chin J Integr Chin West Med 12: 170–171.
10. Zhang P, Wang HW, Lu HJ (1996) Ai-Lin I treated 72 cases of acute promyelocytic leukemia. Chin J Hematol 17: 58–62.
11. Niu C, Yan H, Yu T, Sun HP, Liu JX, et al. (1999) Studies on treatment of acute promyelocytic leukemia with arsenic trioxide: remission induction, follow-up, and molecular monitoring in 11 newly diagnosed and 47 relapsed acute promyelocytic leukemia patients. Blood 94: 3315–3324.
12. Shigeno K, Naito K, Sahara N, Kobayashi M, Nakamura S, et al. (2005) Arsenic trioxide therapy in relapsed or refractory Japanese patients with acute promyelocytic leukemia: updated outcomes of the phase II study and postremission therapies. Int J Hematol 82: 224–229.
13. Soignet SL, Frankel SR, Duerer DS, Tallman MS, Kantarjian H, et al. (2001) United States multicenter study of arsenic trioxide in relapsed acute promyelocytic leukemia. J Clin Oncol 19: 3352–3360.
14. Zheng PZ, Wang KK, Zhang QY, Huang QH, Du YZ, et al. (2005) System analysis of transcriptome and proteome in retinoic acid/arsenic trioxide-induced cell differentiation/apoptosis of promyelocytic leukemia. Proc Natl Acad Sci USA 102: 7653–7658.
15. Gianni M, Koken MH, Chellib-Aliax MK, Benoit G, Lanotte M, et al. (1998) Combined arsenic and retinoic acid treatment enhances differentiation and apoptosis in arsenic-resistant NB4 cells. Blood 91: 4300–4310.
16. Jing Y, Wang L, Xia L, Chen GQ, Chen Z, et al. (2003) Combined effect of all-trans retinoic acid and arsenic trioxide in acute promyelocytic leukemia cells in vitro and in vivo. Blood 97: 264–269.
17. Lallemand-Breitenbach V, Guillemin MC, Janin A, Daniel MT, Doges L, et al. (1999) Retinoic acid and arsenic synergies to eradicate leukemia cells in a mouse model of acute promyelocytic leukemia. J Exp Med 189: 1043–1052.
18. Shen ZX, Shi ZZ, Fang J, Gu BW, Li JY, et al. (2004) All-trans retinoic acid/As2O3 combination yields a high quality remission and survival in newly diagnosed acute promyelocytic leukemia. Proc Natl Acad Sci USA 101: 3328–3333.
19. Aribi A, Kantarjian HM, Estey EH, Koller CA, Thomas DA, et al. (2007) Combination therapy with arsenic trioxide, all-trans retinoic acid, and gentuzumab ozogamicin in recurrent acute promyelocytic leukemia. Cancer 109: 1355–1359.
20. Estey E, Garcia-Manero G, Ferrajoli A, Faderl S, Verstovsek S, et al. (2006) Use of all-trans retinoic acid plus arsenic trioxide as an alternative to chemotherapy in untreated acute promyelocytic leukemia. Blood 107: 3469–3473.
21. Wang G, Li W, Cui J, Gao S, Xiao G, et al. (2004) An efficient therapeutic approach to patients with acute promyelocytic leukemia using a combination of arsenic trioxide with low-dose all-trans retinoic acid. Hematol Oncol 22: 63–71.
22. Li X, Sun WJ, Li ZJ, Zhao YZ, Li YT, et al. (2007) A survival study and prognostic factors analysis on acute promyelocytic leukemia at a single center. Leuk Res 31: 765–771.
23. Hu J, Liu YF, Wu CF, Xu F, Shen ZX, et al. (2009) Long-term efficacy and safety of all-trans retinoic acid/arsenic trioxide-based therapy in newly diagnosed acute promyelocytic leukemia. Proc Natl Acad Sci U S A 106 (9): 3342–3347.
24. Gore SD, Gojo I, Sekeres MA, Morris L, Devetten M, et al. (2010) Single cycle of arsenic trioxide-based consolidation chemotherapy spares anthracycline exposure in the primary management of acute promyelocytic leukemia. J Clin Oncol 28: 1047–1053.
25. Powell BL, Moser B, Stock W, Gallagher RE, Willman CL, et al. (2010) Arsenic trioxide improves event-free and overall survival for adults with acute promyelocytic leukemia: North American Leukemia Intergroup Study C9710. J Clin Oncol 28: 3751–3757.
26. Quezada G, Kopp L, Estey E, Wells RJ (2008) All-trans-retinoic acid and arsenic trioxide as initial therapy for acute promyelocytic leukemia. Pediatr Blood Cancer 51: 135–133.
27. Mathews V, George R, Lakhmi KM, Viswanadhya A, Bajaj A, et al. (2006) Single-agent arsenic trioxide in the treatment of newly diagnosed acute promyelocytic leukemia: Durable remissions with minimal toxicity. Blood 107: 2627–2632.
28. Ghalavandzadeh A, Alimoghadam K, Ghaffari SH, Rostami S, Jahan M, et al. (2006) Treatment of acute promyelocytic leukemia with arsenic trioxide without ATRA and/or chemotherapy. Ann Oncol 17: 131–134.
29. Dai CW, Zhang GS, Shen JK, Zheng WL, Pei MF, et al. (2009) Use of all-trans retinoic acid in combination with arsenic trioxide for remission induction in patients with newly diagnosed acute promyelocytic leukemia and for consolidation/maintenance in CR patients. Acta Hematol 121 (1): 1–8.
30. Courte SE, Othus M, Powell B, Willman CL, Stock W, et al. (2014) Arsenic trioxide during consolidation for patients with previously untreated low/intermediate risk acute promyelocytic leukemia may eliminate the need for maintenance therapy. Br J Haematol doi: 10.1111/bjh.12775.
31. Asou N, Kishimoto Y, Kiya H, Okada M, Kawai Y, et al. (2007) A randomized study with or without intensified maintenance chemotherapy in patients with acute promyelocytic leukemia who have become negative for PML-RARalpha transcript after consolidation therapy: the Japan Adult Leukemia Study Group (JALSG) APL97 study. Blood 110(1):59–66.
32. Au WY, Chim CS, Lie AK, Liang R, KwongYL (2002) Combined arsenic and retinoic acid in acute promyelocytic leukemia: treatment results and prognostic factor analysis from the North American Intergroup study (JALSG) APL97 study. Blood 110(1):59–66.
33. Grigg A, Kimble R, Szer J (2003) Prolonged molecular remission after arsenic trioxide and all-trans retinoic acid for acute promyelocytic leukemia relapsed after allogeneic stem cell transplantation. Leukemia 17: 1916–1917.

34. Galimberti S, Papineschi F, Carmignani A, Testi R, Fazzi R, et al. (1999) Arsenic and all-trans retinoic acid as induction therapy before autograft in a case of relapsed resistant secondary acute promyelocytic leukemia. Bone Marrow Transplant 24: 345–348.

35. Rock N, Mattiello V, Judas C, Huezo-Diaz P, Bourquin JP, et al. (2014) Treatment of an acute promyelocytic leukemia relapse using arsenic trioxide and all-trans-retinoic in a 6-year-old child. Pediatr Hematol Oncol 31(2):143–148.

36. Raffoux E, Rousselot P, Poupon J, Daniel MT, Cassinat B, et al. (2003) Combined treatment with arsenic trioxide and all-trans-retinoic acid in patients with relapsed acute promyelocytic leukemia. J Clin Oncol 21: 2396–2404.

37. Zhang Y, Zhang Z, Li J, Li L, Han X, et al. (2013) Long-term efficacy and safety of arsenic trioxide for first-line treatment of elderly patients with newly diagnosed acute promyelocytic leukemia. Cancer 119(1):115–125.

38. Lo-Coco F, Avvisati G, Vignetti M, Thiede C, Orlando SM, et al. (2013) Retinoic acid and arsenic trioxide for acute promyelocytic leukemia. N Engl J Med 369(2):111–121.

39. Iland HJ, Bradstock K, Supple SG, Catalano A, Collins M, et al. (2012) All-trans-retinoic acid, idarubicin, and IV arsenic trioxide as initial therapy in acute promyelocytic leukemia (APML4). Blood 120(3):1570–1580.

40. Avvisati G, Lo-Coco F, Paoloni FP, Petri MC, Diverio D, et al. (2011) AIDA 09/3 protocol for newly diagnosed acute promyelocytic leukemia: very long-term results and role of maintenance. Blood 117(18):4716–4725.

41. Sanz MA, Montesinos P, Rayón C, Holowiecka A, de la Serna J, et al. (2010) Risk-adapted treatment of acute promyelocytic leukemia based on all-trans retinoic acid and anthracycline with addition of cytarabine in consolidation therapy for high-risk patients: further improvements in treatment outcome. Blood 115(25): 5137–5146.

42. Lengfelder E, Haferlach C, Saussele S, Haferlach T, Schultheis B, et al. (2009) High dose ara-C in the treatment of newly diagnosed acute promyelocytic leukemia: long-term results of the German AMLCG. Leukemia 23(12):2248–2258.

43. Dos Santos GA, Kats L, Pandolfi PP (2013) Synergy against PML-RARα: targeting transcription, proteolysis, differentiation, and self-renewal in acute promyelocytic leukemia. J Exp Med 210(13):2793–2802.