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Current Study of APL Treatment in China

Dao-Pei Lu, Qin Wang

Peking University Institute of Hematology, Peking University People's Hospital, China

Abstract

Natural As_4S_4 and As_2S_3 are major components in realgar and orpiment, which also contain small amount of As_2O_3 and other toxic materials. Liu YF showed with us that NB4 cell line and its mice model were more sensitive to As4S4 than to As_2O_3 and As_2S_3. That was in agreement with the clinical data available from China. Wang FR and we proved in mice and in clinical pharmaceutical study that ground *Seman platycladi* as an excipient can appreciably increase the blood level of arsenic when taken P.O. together with As_4S_4. Our clinical study with 130 patients with t(15;17) APL showed both As_4S_4 and As_2S_3 alone were very effective in CR induction, including cytogenetic remission and PML-RAR reversion, yet the relapse rate was higher in the group treated by As_4S_4 alone than in the group treated together with other autileukemic agents, either together or in sequence. In the later group, LFS for 1 and 6 years was 96.7% and 87.4%, and the over-all survival for 1 and 6 years was 98.9% and 93.9%. High dose chemotherapy was proven to be not only unsafe but also unnecessary. In newly diagnosed APL patients with neutropenia, CR could be safely achieved by As_4S_4 P.O. and/with ATRA. Chemotherapy was then followed. In patients with leukocytosis, chemotherapeutic agents such as hydroxyurea or harringtonine was added at the beginning and then followed by anthracycline and asparaginase. The reasons of APL relapse after patients receiving As_4S_4 P.O. were: ① emergence of a resistant cell clone showing additional chromosomal abnormalities other than t(15;17); ② decrease of blood As level. The resistant APL case could respond favorably to combination of As_4S_4 P.O. together with ATRA. The blood level of As could be raised by modification of the preparation under investigation. Allogeneic stem cell transplantation (allo-SCT) is the last option in relapsed patients, became autologous stem cell transplantation (ASCT) has a significantly better outcome. In a few hospitals ASCT was performed early in the CR1. In rare cares, patients were referred with intra-cranial hemorrhage. CT or EMR scan was done urgently at this instance. Cranial surgery carried out without a minute of delay resulted in the rescue and long survival of patient, while delay of surgery had resulted in irreversible damage of Pons/ Medulla oblongata and ended in death. As_2O_3 IV had been used with success in mentally unclear patients not suffering from intra-cranial hemorrhage. Although no parallel clinical trial has been done to compare the efficacy of As_2O_3 and As_4S_4, yet the fact was it was easier for doctors and patients to carry out and receive maintenance treatment with oral As_4S_4 rather than with As_2O_3. According to the data and principles above, most of the t(15;17) APL patients can be cured.

Acute promyelocytic leukemia (APL) used to be one of critical/fulminant human malignancies, usually accompanied with a life-threatening hemorrhagic diathesis. Malignant promyelocytes grow out of control and fail to differentiate into normal mature granulocytes. Typical promyelocytes possess a unique chromosome translocation t(15;17), resulting in a chimeric gene, promyelocytic leukemia-retinoic acid receptor-α (PML-RARα). In China the treatment of APL has been extensively studied and achieved prominent results. All-trans retinoic acid (ATRA) and arsenic compounds dramatically and specifically improved the clinical course and the long-term outcome of patients with APL.

Natural tetra-arsenic tetra-sulfide (As_4S_4) and diarsenic trisulfide (As_2S_3) are major components in realgar and orpiment, which also contain small amount of arsenic trioxide (As_2O_3) and varieties of other toxic materials. Realgar and orpiment have been used as traditional

medicines in China for more than 1500 years. Either externally or by mouth, realgar and orpiment were administered either to treat or to prevent from some diseases, and regarded as having mild toxicity. Medical use of mined realgar without being processed is however toxic because of appreciable amounts of As_2O_3 as well as calcium and magnesium arsenites in unrefined realgar. Even though realgar was used popularly in China, it has been employed with a number of herb drugs containing hundreds of components. Neither As_4S_4 nor other arsenic sulfides such as As_2S_3 has been described to be effective against any malignancy by being used alone before our report. In recent years, we have undertaken a series of studies in vitro and in vivo, and demonstrated that both As_4S_4 and As_2S_3 are effective in the treatment of APL.

1. Experimental and Animal Studies of As_4S_4

Study by Liu YF et al showed that As_4S_4 and As_2S_3 had an inhibitory effect on cell viability of NB4 cells in the time- and dosage-dependent manner. The half inhibitory rate (IC_{50}) of As_2S_3 was 1.19×10^{-6} mol/L. As_4S_4 exerted a more prominent inhibitory effect than that of As_2S_3 (P=0.001), with an IC_{50} of 2.86×10^{-7} mol/L. As_4S_4 and As_2S_3 promoted apoptosis in NB4 cell, including typical morphologic changes, increase in subdiploid population and DNA ladder formation on gel electrophoresis. The apoptosis of NB4 cells was associated with the activation of caspase-3. Arrest of cell cycle in G2/M phases was also observed in NB4 cells. In the therapeutic trial of SCID mice model transplanted with human APL-ascites, survival of the experimental animal was 16.13±2.33 days in the control group, 24.25±3.41 days in the As_4S_4 group and 24.05±3.54 days in the As_2S_3 group (vs control group P<0.01). The survival of SCID mice bearing human APL ascites was extended by treatment of As_4S_4 and As_2S_3. These findings suggest that NB4 cell line and its mice model were sensitive to As_4S_4 and As_2S_3. It was in agreement with the clinical data available from China.

In another study by Wang FR et al, two kinds of As_4S_4 preparations, As_4S_4 alone and As_4S_4 with ground *Seman platycladi* as an excipient, were used to study the difference of As_4S_4 absorption through gastrointestinal track in two groups of mice. The two groups were given the same dose of 2,500 mg/kg As_4S_4. When As_4S_4 was administrated with *Seman platycladi*, the blood maximal concentration (C_{max}) of arsenic was increased to 399.8±105.1μg/L from 329.1±110.7μg/L (group with As_4S_4 alone), and the time to peak concentration (T_{peak}) was delayed, but the elimination half-life [$t_{1/2}$(ke)] was prolonged.

2. Clinical Pharmacokinetic Study of As_4S_4

Wang FR's and our clinical pharmacokinetic study of As_4S_4 with ground *Seman platycladi* was taken in 18 voluntary APL patients in HCR at a single dosage of 60 mg/kg As_4S_4. The results showed that blood arsenic could be detected 30 minutes after oral administration of As_4S_4, whereas it reached the peak level with T_{peak} 2~3 hours and C_{max} 20~28μg/L. The arsenic excretion in urine can be detected as early as 4 hours after drug administration. The amount of arsenic excretion within the first 24 hours accounted for 50% of the total arsenic excretion during the first 96 hours, which accumulated to 3479 2338.5 microgram in total.

3. Clinical Study of As_4S_4

Our clinical study with 130 patients with t(15;17) APL showed both As_4S_4 and As_2S_3 alone were very effective in CR induction, including cytogenetic remission and PML-RAR reversion.

As_4S_4 was given to 129 patients with APL. Nineteen were newly diagnosed, 7 in the first relapse, and 103 in hematological complete remission (HCR). HCR was achieved in all newly diagnosed and hematological relapsed cases. Of 16 newly diagnosed patients with available cytogenetic/molecular analyses, 14 cases attained cytogenetic and molecular CR. Cytogenetic/molecular CR was also obtained in 5 of 7 hematological relapsed cases. In HCR group, 35 out of 44 patients with PML-RARα positive at baseline were rendered to negative. In newly diagnosed group, the estimated leukemia-free survival (LFS) for 1 and 3 years were 86.1% and 76.6%, and the over-all survival (OS) for 1 and 3 years was 93.3% and 70.0%. In the HCR group, LFS for 1 and 6 years was 96.7% and 87.4%, and OS for 1 and 6 years was 98.9% and 93.9%. Yet the relapse rate was higher in the newly diagnosed group treated by As_4S_4 alone than in the HCR group treated together with other autileukemic agents, either together or in sequence (18.7% vs 8.8%).

As_4S_4 treatment was well tolerated with only moderate side effects in a minority of patients or in isolated cases, including asymptomatic prolongation of corrected QT interval, transient liver enzyme elevation, skin rashes and mild gastrointestinal discomfort. There was neither myelosuppression nor appreciable long-term side effects. As_4S_4 was given safely to newly diagnosed APL patients with severe neutropenia. CR could be safely achieved by As_4S_4 P.O. and/with ATRA. Chemotherapy was then followed. Leukocytosis was absent when patients achieved HCR. However, mild and transient leukocytosis was detected in some of newly diagnosed patients during remission induction period. For patients with leukocytosis chemotherapeutic agents such as hydroxyurea or harringtonine were added at the beginning. Some of patients resolved spontaneously without administration of cytotoxic drugs.

In the course of this current study, we used synthetic and chemically pure As_2S_3 to treat a single patient with newly diagnosed APL. After monotherapy with As_2S_3, this patient entered hematological CR in 38 days and molecular CR in 128 days. It thus reveals that this form of arsenic sulfide is also an effective and safe therapy for APL.

4. Issues in the Management of APL

4.1. Induction Therapy

Merit of ATRA resides in that along with HCR of APL there is improvement of coagulopathy without repression of bone marrow. Advantage of arsenic is not only to improve coagulopathy but also to achieve cytogenetic normalization and molecular CR that is usually difficult to be attained in the monotherapy of ATRA. Therefore, we suggest that it would be better choice to combine ATRA with arsenic, including either low dose of As_2O_3 or oral As_4S_4, in the initial induction therapy until HCR is attained. When the white blood cell count began to go up during induction, hydroxyurea or low dose of harringtonine (2 mg/day) was administered.

4.2. Post-remission Therapy

After HCR, intermittent chemotherapy seems to be essential. However, it is neither safe nor necessary to expose patients to the risk of high dose of chemotherapy. Moderate or even low doses of cytotoxic agents, including harringtonine (2 mg/day×10) with asparaginase (10,000 U, q3d×12), or mitexantrone (2 mg/day) or anthracycline, is followed intravenously. Then intrathecal treatment of MTX is administered to prevent from extramedullary relapse.

The reasons of APL relapse after patients receiving As_4S_4 P.O. include: ① emergence of a resistant cell clone showing additional chromosomal abnormalities other than t(15;17); ② decrease of blood arsenic level. The resistant APL case could respond favorably to combination of As_4S_4 P.O. together with ATRA.

4.3. Maintenance Therapy

Our experience with oral As_4S_4 suggest that As_4S_4 could provide APL patients not only cost and quality-of-life benefit but also easy access to consolidation and maintenance therapy. For patients after HCR, the identical daily dose of 50 mg/kg As_4S_4, which was divided into four dosages (approximately 750 mg qid), was given on a treatment schedule of 2 weeks on and 2 weeks off in the first year. Thereafter the treatment break was increased to one month within four years. The therapy was discontinued in the fifth year.

Parallel clinical trial has not been done to compare the efficacy of As_2O_3 and As_4S_4. However, the fact was that lethal liver and cardiac toxicity in patients during induction treatment with As_2O_3 had been reported. It was easier for doctors and patients to carry out and receive maintenance treatment with oral As_4S_4 rather than with As_2O_3.

4.4. Bone Marrow Transplantation

Allogeneic stem cell transplantation (allo-SCT) is the last option in relapsed patients, because autologous stem cell transplantation (ASCT) has a significantly better outcome. Our previous study on evaluating treatment efficacy of SCT and As_4S_4 therapy showed that there was no statistical difference between the outcome of all-SCT and ASCT in the 3-year LFS or OS, but the 1-year LFS or OS was better in ASCT. Noticeably, As_4S_4 therapy was proven to be much safer and more efficient together with chemotherapy than either all-SCT or ASCT. In a few hospitals ASCT was routinely performed early in the CR1.

4.5. Treatment of Severe Complication

In rare cares, patients were referred with intra-cranial hemorrhage. CT or EMR scan was done urgently at this instance. Cranial surgery carried out without a minute of delay resulted in the rescue and long survival of patient, while delay of surgery had resulted in irreversible damage of Pons/Medulla oblongata and ended in death. As_2O_3 IV had also been used with success in mentally unclear patients but not suffering from intra-cranial hemorrhage.

5. Summary

Chemotherapy, ATRA and arsenics have been proven to be the three milestones in the treatment of t(15;17) APL. Oral As_4S_4 is not only effective but also safer in remission induction as well as maintenance treatment. Most APL patients can be cured with As_4S_4 as the main drug together with ATRA and chemotherapy. It is not necessary to expose the patients with APL to high risks anymore. However, in relapsed patients stem cell transplantation still remains to be the last option.