Might arsenic trioxide be useful in the treatment of advanced myelodysplastic syndromes?

Failure of conventional therapies is dramatic in patients with advanced myelodysplastic syndromes (MDS), thus therapeutic approaches that increase the apoptotic rate of myelodysplastic syndrome (MDS) cells might be advantageous. We studied the behavior of cells exposed to As_2O_3 *in vitro* from patients with advanced MDS. Our data show that therapeutically useful concentrations of As_2O_3 increase the rate of apoptosis in short-term cultures. Moreover, pre-treatment with GM-CSF increased the sensitivity of the cells to the effect of As_2O_3.

Sir,

The search for therapy of myelodysplastic syndromes has so far been unsuccessful. Single agent chemotherapy as well as acute myeloid leukemia (AML)-type chemotherapy, alone or in combination with human growth factor (HGF), have not shown a clear beneficial effect on survival in MDS or in MDS-related AML.

Morphologic features of apoptosis are easily demonstrable in MDS cell populations.[1] Moreover, data about fas expression in $CD34^+$ MDS cells show that the myeloblast populations that emerge with disease progression are more resistant to *fas*-induced cell death.[2] Thus, it is possible to hypothesize that, in advanced phases of MDS, an increase in the apoptotic rate of MDS cell populations might be advantageous.

Recently, much attention has been drawn to the use of As_2O_3 as a useful agent in the treatment of AML-M3 that has become resistant to standard retinoic acid treatment.[3] In a patient with retinoic-resistant AML-M3 treated with arsenic trioxide we observed a very good correspondence between the levels of drug-induced apoptosis in affected cells *in vivo* and that observed *in vitro* in liquid culture. This correspondence was maintained during progression of the disease (4 and AD, personal communication).

In the present study, we collected blood samples from 14 patients with the diagnoses shown in Table 1. Bone marrow or peripheral mononuclear cells were seeded in 6-well plates (250,000 cells/mL IMDM/10% FCS). After 24 hours, As_2O_3 was added at concentrations of 0.1, 0.5, 1 and 2 μM. After 5 days in culture, culture aliquots were stained with propidium iodide and analyzed by FACS using Lysis II software. The percentage of cells in the hypodiploid peak was used to assess the percentage of apoptotic cells. A minimum of 10,000 events were taken for each sample.

The result obtained show that micromolar concentrations of As_2O_3 were effective at inducing apoptosis in 7/11 advanced MDS cases. No apoptosis was induced in cases of AML-M0 and Ph+ALL.

We also observed that the drug was more effective in inducing apoptosis when the initial spontaneous levels of apoptosis were low, suggesting that the drug might preferentially act on cycling cells. To test this hypothesis we repeated the experiments adding the

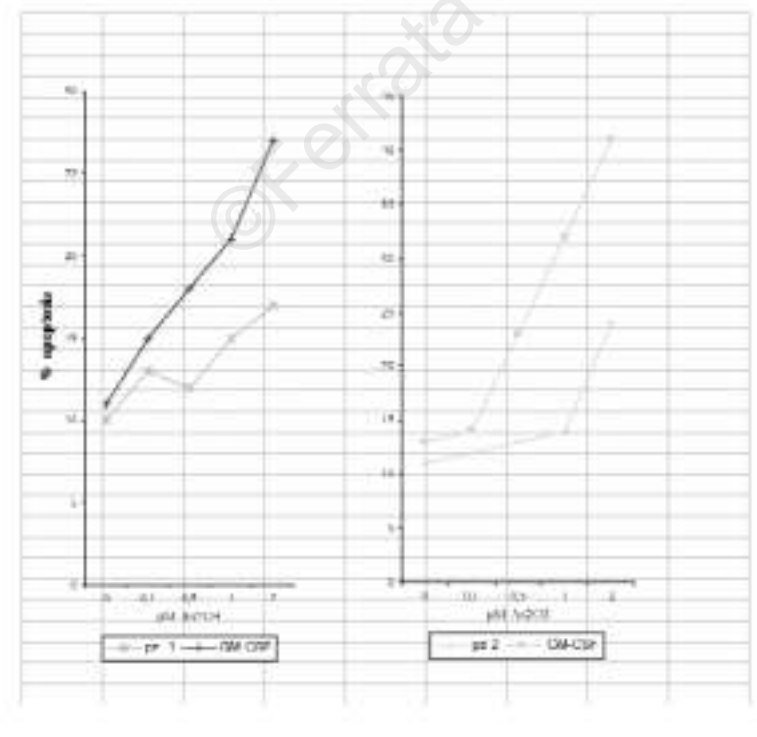

Figure 1.

Table 1.

Diagnosis	Number of cases	Response to As_2O_3	Response to +GM-CSF
RAEB-t	3	2/3	ND
AML-MDS	8	5/8	4/8
AML-M3	1	1/1	ND
AML-M0	1	0/1	0/1
CMML	1	0/1	1/1

As_2O_3 24 hours after exposing the cells to GM-CSF. In 5/8 cases the addition of GM-CSF made the cells more sensitive to the apoptotic effects of As_2O_3, particularly at concentrations as low as those known to be effective *in vivo*. Representative experiments are shown in Figure 1.

Two interesting observations can be drawn from our data. The initial levels of apoptosis in our experiments are strikingly similar to those observed in vivo (14% vs 10-15% in ref. #2). Moreover, a relevant proportion of cells from advanced MDS cases seem to respond in vitro to the anti-apoptotic effect of HGF. The role played by the process of cell death in the pathogenesis of MDS is still a matter of debate.[4] We show here that a combination of a pro-apoptotic agent with a differentiative stimulus seems to be able to overcome the relative resistance to apoptosis documented in advanced MDS cases.

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Key words

Arsenic trioxide, myelodysplastic syndromes.

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Epoch-cyclosporin A induces high response rates in relapsed lymphomas

We describe the results concerning tolerance, response and survival of a program of rescue therapy for primary refractory and relapsed lymphoma combining cyclophosphamide and prednisone with a 96-hour infusion of etoposide, vincristine, and doxorubicin (EPOCH) with cyclosporin A (CsA).

Sir,

The prognosis of patients with refractory or relapsed lymphoma is very poor in spite of aggressive salvage treatments.[1] Multiple molecular mechanisms involved in drug resistance have been elucidated.[2] It is possible to circumvent some of them by using specific modulators.[3-5] Cyclosporin A can effectively modulate multidrug resistance mediated by P-glycoprotein[4] and other mechanisms.[6] Otherwise, the administration of chemotherapeutic agents as a continuous infusion can circumvent the resistance of patients treated with the same drugs administered in bolus.[7]

In August 1994, we began a program of rescue therapy combining EPOCH infusional chemotherapy[7] with CsA. An intravenous (i.v.) loading dose of CsA 5 mg/kg was given over 2 hours on day 1, followed by 16 mg/kg/d as i.v.-continuous infusion on days 1 to 4. Simultaneously with the CsA continuous infusion of etoposide, vincristine, and doxorubicin were all administered on days 1 though 4. Finally, cyclophosphamide (i.v. bolus on day 6) and prednisone (orally on days 1 through 6) were given. Treatments were repeated every 4 weeks. When possible, patients achieving a response with CsA+EPOCH chemotherapy were consolidated either with high dose chemotherapy and/or radiotherapy.

The characteristics of the 13 patients treated in this way are shown in Table 1.

A total of 51 cycles of CsA+EPOCH were administered. The median number of treatment cycles per patient was two, the range 1-15. Five patients (38%) experienced grade IV leukopenia. Infectious episodes occurred in three patients, and two patients died from these. Other mild toxic events, such as grade II mucositis, fluid retention, hyperbilirubinaemia, pain and central nervous system toxicity were frequently observed. Dose adjustments were required in 6 patients (46%).

The CsA steady-state concentrations showed a high interpatient variability (median 2,117 ng/mL, range 902-2,906). As CsA delays the clearance of some chemotherapeutic agents resulting in enhanced serum drug levels,[8,9] a forty-percent dose reduction with respect to the original EPOCH (without CsA)[7] was initially employed to compensate for this pharmacokinetic interaction and led to a feasible schedule.

There were six objective remissions (46%; 95% C.I.: 19 to 75%): one partial and five complete remissions. The durations of the response were 6 months for the partial and 5, 8, 8, 14, and 34+ months for the complete ones. No responses were seen in the six patients who had failed to achieve a response with their pri-