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Autologous Bone Marrow Transplantation in Acute Leukemia and Non-Hodgkin's Lymphoma: A Phase I Study of 4-Hydroperoxycyclophosphamide (4HC) Incubation of Marrow prior to Cryopreservation *

H. Kaizer, P. Tutschka, R. Stuart, M. Korbling, H. Braine, R. Saral, M. Colvin, and G. Santos

The presence of occult tumor cells in remission marrow is the initial obstacle to the use of autologous bone marrow transplantation in acute leukemia. In vitro treatment of tumor-marrow mixture, with heterologous antiserum and complement, has been shown to be capable of eliminating all clonogenic tumor in a number of animal models, and this approach forms the basis for a number of ongoing clinical trials [1]. An alternative procedure, utilizing in vitro drug treatment of marrow, was first investigated at The Johns Hopkins Oncology Center using 4-hydroperoxycyclophosphamide (4HC). In aqueous solution, this congener of cyclophosphamide (CY) has the same alkylating and cytotoxic effects and the same immunoreactive products as the microsomally activated parent compound.

The initial studies of this approach utilized a model of acute myelogenous leukemia (AML) in the Lewis-Brown Norway (LBN) hydrid rat and involved the inoculation of lethally irradiated rats with marrow tumor cell mixtures which had been treated with various doses of 4HC. These experiments revealed a dose-dependent clearing of tumor cells [2]. Since the LBN-AML model displays an unusual sensitivity to the anti-tumor effects of CY, a model of AML in the Wistar-Furth (WFU) rat, which is not as sensitive to CY as the LBN model, has recently been studied. Despite the relative in vivo insensitivity of the WFU model, similar results have been obtained in treating marrow tumor cell mixtures with varying concentrations of 4HC. Complete elimination of tumor cells has been achieved at concentrations of 60–80 n*M*/ml (18–24 μg/ml) of 4HC. Increasing the concentration of 4HC beyond 80 n*M*/ml resulted in the death of a high fraction of the rats due to marrow failure [3].

We have begun a two-phase study for patients with acute leukemia, either lymphoblastic (ALL) or nonlymphoblastic (ANLL), using 4HC treatment of marrow. The goal of the first phase of this study, which is still ongoing, is to determine the maximal concentration of 4HC that can be used for in vitro treatment and still achieve hematologic recovery. The goal the second phase of the study will be to determine if treatment with 4HC will eliminate all clonogenic tumor.

Thus far, a total of 20 patients with acute leukemia and five patients with non-Hodgkin's lymphoma have been treated on this study as shown in Table 1. This is a dose escalation study. Remission marrow is harvested and aliquoted into a treated and a reserve fraction. The latter is intended for use only if the patient shows no evidence of hematologic recovery after reinfusion of the treated marrow. The pretransplant cytoreductive regimen for ALL and NHL involves CY and total body irradiation. This differs from the regimen used in ANLL which uses CY and busulfan. Both these preparative regimens have been previously described [4]. Although the morbidity of the procedure is significant, only 1 of the 25 patients has had a transplant-related death. All of the other patients have been dis-

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Conc. of 4HC	ALL	ANLL	NHL	% inhibition of CFU-C
40 μg/ml	4	0	1	73
60 μg/ml	2	2	3	85
80 μg/ml	3	3	0	97
100 μg/ml	5	1	1	100

Table 1. Phase I-4HC study patients to July 1, 1982

charged from the hospital in good clinical condition (the average post-transplant stay has been 35 days) and have been able to resume normal activity. Table 1 also shows the degree of inhibition of granulocyte and macrophage colony forming cells (CFU-C) observed as a consequence of 4HC incubation. Despite the high degree of inhibition, essentially no CFU-C detected after incubation at the higher 4HC concentrations, hematologic recovery has been satisfactory in all patients except the one who died of a transplant-related complication (veno-occlusive disease of the liver) too early to evaluate for hematologic recovery. Among the 24 patients evaluable for tumor status, nine remain in remission. Most of those have relatively short observation times, although one ALL and one ANLL patient have been in complete remission for over 15 months. While the disease-free status of those patients contributes to our evaluation of the therapeutic efficacy of 4HC incubation, the ultimate test will be on data collected in the phase II part of the study. This cannot be instituted until the maximal concentration of 4HC is established.

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