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Autologous Bone Marrow Transplantation Following High-dose Chemotherapy with Cyclophosphamide, BCNU and VP-16 in Small Cell Carcinoma of the Lung and a Review of Current Literature

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Abstract—*Four patients with limited disease small cell carcinoma of the lung were treated with high-dose cyclophosphamide* (120 mg/kg *over days* 1 *and* 2), *BCNU* (400 mg/m^2 *over days* 1 *and* 2) *and VP-16* (200 mg *days* 1–5) *used as intensification treatment after conventional chemotherapy. To ameliorate hematopoietic toxicity, autologous bone marrow cells collected and cryopreserved prior to treatment were reinfused on day* 8. *In three patients clinical response was evaluable. Two achieved a complete remission: one died without evidence of tumor after* 3 months; *the other had a regional relapse after* 6 months. *One patient who had progression of disease on conventional chemotherapy was refractory to high-dose treatment. Three patients developed diffuse interstitial pneumonitis* 3 weeks *after treatment and two died of respiratory failure. High-dose intensification chemotherapy with autologous marrow reinfusion may complement the effects of standard combination chemotherapy in small cell carcinoma of the lung. The current status of this approach is reviewed.*

INTRODUCTION

LUNG CANCER is responsible for one-quarter of cancer-related deaths in the United States [1]. Approximately 25% of all lung cancers are of the small cell variety [2]. Within the past decade a considerable amount of investigation in small cell carcinoma of the lung (SCC) has shown a propensity for early metastases but also a responsiveness to multiple chemotherapeutic agents. Combination chemotherapy has become the standard treatment for SCC and, with currently used regimens, over half of the patients can be expected to achieve complete remission [3]. However, despite a good response to chemotherapy, the vast majority of patients relapse within a year, and most patients die of recurrent disease within 18 months [4]. Experimental animal tumor models and clinical observations suggest a steep dose–response relationship for tumors sensitive to chemotherapeutic agents [5]. The high order of responsiveness of SCC to a variety of chemotherapeutic agents suggests an unusual sensitivity of tumor cells which might be further enhanced by intensification of dose. The limiting toxicity of most agents useful in the treatment of SCC is myelosuppression. Infusion of autologous marrow cells may accelerate the recovery of blood counts by several days and allow for intensification of the dose of myelosuppressive agents [6].

The present report gives the results of a pilot trial of high-dose chemotherapy with autologous marrow reinfusion in SCC as intensification therapy after conventional chemotherapy.

MATERIALS AND METHODS

Four patients (3 male, 1 female) with a median age of 53 (range 41–61) yr entered the study after giving informed consent. All had limited disease at presentation on staging by physician examination, blood chemistries, chest X-ray, liver and bone scans, head CT, and bone marrow aspirate

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and biopsy. All received an initial regimen consisting of cyclophosphamide 700 mg/m^2 i.v. day 1, adriamycin 45 mg/m^2 day 1, vincristine 1 mg/m^2 i.v. day 1 and methotrexate 3 g/m^2 i.v. days 8 and 15, followed in 24 hr by intravenous leucovorin rescue (CAV-HDMTX). The regimen was repeated every 3 weeks. Prior to initiation of chemotherapy, two patients received low-dose mediastinal irradiation (900 and 1500 rad) to control symptoms of superior vena cava obstruction. After four cycles of CAV-HDMTX, two patients achieved a partial clinical remission. One patient had a complete clinical response, but the CEA, initially elevated at 138 ng/ml, remained elevated at 42 ng/ml. After five cycles of CAV-HDMTX, the fourth patient had progressive disease in the chest and evidence of new CNS metastasis by CT scan. He received 1800 rad of cranial irradiation before high-dose chemotherapy. The patient characteristics are summarized in Table 1.

All patients had a bone marrow negative for tumor at diagnosis. After induction chemotherapy with CAV-HDMTX, a repeat bone marrow aspirate and biopsy were performed. A bone marrow negative for tumor cells was required for subsequent high-dose chemotherapy with autologous bone marrow reinfusion. After harvesting of bone marrow under general anesthesia, the patients received a single course of high-dose chemotherapy with cyclophosphamide 120 mg/kg i.v. in divided doses over days 1 and 2, BCNU 400 mg/m^2 in divided doses over days 1 and 2, and VP-16 200 mg/m^2 i.v. on days 1–5 (HDCBV). The marrow was reinfused on day 8. Bone marrow was aspirated from the anterior and posterior iliac crests with Rosenthal needles, with the patient under general anesthesia. A mean of 2×10^6 nucleated cells was obtained. Clotting of the marrow was prevented by aspirating the marrow cells into anticoagulant citrate dextrose solution, USP, a formula containing 50 units/ml heparin sodium (preservative free). The leukocyte portion of the marrow was concentrated by dextran sedimentation, washed and resuspended in Hanks' balanced salt solution and 25% human AB+ serum. DMSO was added to a final concentation of 10%. Utilizing a planar nitrogen-freezing apparatus, the marrow was placed in 100-ml Hemoflex bags, frozen at -1°C/min and stored in the vapor phase of liquid nitrogen. Prior to infusion, the marrow was thawed at 40°C (no DNase was used).

Tumor response was evaluated 1 month after high-dose intensification. Complete response was defined as disappearance of all signs of disease for at least 4 weeks. A partial response was defined as 50% or greater reduction in tumor size. Duration of response was measured from the time of high-dose chemotherapy to the time of progression. No maintenance chemotherapy was given.

RESULTS

After HDCBV with autologous marrow infusion, all patients went through a period of pancytopenia. The median time from high-dose intensification to recovery from neutropenia to over 500 neutrophils/mm^3 was 29 (range 22–42) days. Platelet counts over 50,000/mm^3 were reached after 37, 47 and 66 days in three evaluable patients. During the period of myelosuppression, one patient developed fever for which no cause was found; another had a Gram-negative sepsis that was successfully treated with antibiotics and granulocyte transfusions.

Three of four patients developed diffuse interstitial pneumonitis 3 weeks after high-dose intensification. The pneumonitis subsided spontaneously within 3 weeks in one patient, but two patients died of respiratory failure after 1 and 3 months. Post-mortem examination of the lungs disclosed diffuse fibrosis and alveolar damage,

Table 1. *Results of intensification with HDCBV in SCC**

Patients	Response to induction	Response to HDCBV	Toxicity of HDCBV	Outcome
A	radiological CR, residual elevation of CEA (?lab PR)	CR, 3+ months	fatal pneumonitis, culture-negative fever	died of respiratory failure; disease-free at autopsy
B	radiological PR	not evaluable	fatal pneumonitis, hepatitis, Gram-neg. sepsis	died of respiratory failure; residual local disease at autopsy
C	radiological PR†	CR, 6 months	pneumonitis, hepatitis	died of local and systemic relapse
D	progressive disease†	progressive disease	—	died of disease

*HDCBV = high-dose cyclophosphamide, BCNU and VP-16.
†Mediastinal irradiation for SCC prior to initial chemotherapy.

both consistent with BCNU toxicity [7]. Hepatotoxicity was the only other significant extramedullary toxicity observed. One patient developed a cholestatic picture with ten-fold elevation of the alkaline phosphatase 2 weeks after HDCBV. He died from pulmonary complications before hepatic recovery. Another patient had a ten-fold elevation of hepatocellular enzymes 3 weeks after HDCBV that normalized after several weeks.

Two responses were seen in three clinically evaluable patients. Patients A and C had complete responses lasting 3 and 6 months respectively. Patient A died from treatment-related toxicity and was found to be tumor-free at autopsy. Patient C had local relapse in the chest and new evidence of ipsilateral supraclavicular node metastasis. At the time of relapse, no tumor was found in the marrow. Patient D, whose tumor was refractory to induction chemotherapy, also failed to respond to the high-dose program and died of progressive disease. One month after HDCBV, a repeat bone marrow biopsy in this patient showed evidence of marrow involvement. Patient B died too early to evaluate a clinical response. At autopsy he was found to have residual tumor in the bronchus. The results of treatment and major toxicities of HDCBV are summarized in Table 1.

DISCUSSION

High-dose chemotherapy still represents a major potential for improving systemic therapy in SCC. The rationale for high-dose chemotherapy is based on experimental *in vivo* systems and on clinical observations in tumors highly responsive to chemotherapeutic agents [6]. In most experimental tumors there is a linear logarithmic relationship between dose and cell kill, e.g. a two-fold increase in dose may lead to a ten-fold increase in cell kill. In one study of SCC the doubling of the dose of cyclophosphamide from 500 to 1000 mg/m^2, in combination with standard doses of BCNU and methotrexate, leads to an increase in complete remissions from 0 to 30% and in overall responses from 45 to 96% [8]. A dose-response relationship in SCC has recently also been suggested for VP-16 [9] but could not be documented for methotrexate [10].

An increase in dose might have a beneficial antitumor effect but is also accompanied by increased toxicity. Autologous marrow support was chosen in our study to ameliorate prolonged myelotoxicity from HDCBV, especially the delayed myelosuppression of BCNU. The exact role of autologous marrow support in high-dose chemotherapy for SCC is investigational. It has been recently demonstrated that cyclophosphamide used as a single agent can be escalated to 7 g/m^2 without using autologous marrow [11]. However, certain agents, such as BCNU, melphalan and dimethylmyleran, are known to be associated with prolonged myelosuppression and autologous marrow reinfusion may be beneficial with these agents.

Reinfusion of autologous marrow cells theoretically poses the risk of seeding viable tumor cells. With monoclonal antibodies tumor cells can be demonstrated in bone marrow aspirates from patients with SCC that are found to be negative by conventional histological methods [12]. However, relapses tend to occur in sites of previous bulk disease, and it is difficult to demonstrate the tumor-forming potential of a small amount of SCC cells that might be present in the reinfused autologous marrow. With progress in methodology, it should be possible in the future to purge autologous marrow cells from contaminating tumor cells by monoclonal antibodies.

Severe pulmonary toxicity was noted in this trial and precluded further accrual. Three patients developed diffuse interstitial pneumonitis. One recovered, but two died from respiratory failure. This high incidence of pulmonary toxicity was unexpected. The clinical course and post-mortem findings suggested that the pneumonitis was related to BCNU. Pathologically, there was no evidence for CMV infection. The incidence of pulmonary toxicity from BCNU is less than 10% with cumulative doses of 400 mg/m^2 [13]. However, these data were obtained in patients with extrapulmonary tumors receiving BCNU as a single drug. History of lung disease and tobacco use, both highly prevalent in patients with SCC, were associated with a significant increase in the pulmonary toxicity of BCNU [13]. Another cause of the high incidence of pneumonitis in our study might have been the concomitant use of cyclophosphamide. Such an unfavorable interaction has been suggested by two other reports [14, 15]. Two series of high-dose chemotherapy in SCC employed a regimen similar to HDCBV used in our study. In one, BCNU was omitted from some patients, and the pulmonary toxicity was not analyzed according to the agents employed [16]. In the other study two of six patients treated with BCNU and cyclophosphamide died of fatal pneumonitis [17].

Our study shows that with high-dose intensification complete remissions can be obtained in patients with only a partial response to a conventional regimen. All four patients had evidence of residual disease after CAV-HDMTX, and two subsequently achieved complete remission by HDCBV. One died in complete remission

after 3 months; the other had evidence of only regional relapse after 6 months.

Several other studies using high-dose chemotherapy in SCC have been published, and the findings are summarized in Table 2. High-dose chemotherapy was employed in patients failing primary treatment as the initial form of chemotherapy or as intensification after conventional chemotherapy. Of 22 patients from five reports who failed prior treatment, four (22%) achieved complete remission by high-dose chemotherapy [16, 18–21]. The duration of complete response reported in two patients was 2 and 3 months respectively [16, 21]. Two groups reported on the use of high-dose chemotherapy in previously untreated patients [22, 23]. Of 39 patients, 21 (44%) achieved complete remission. In one study patients were maintained with combination chemotherapy during remission [23]. Neither the duration of remission or survival in these studies were better than results of recent reports using conventional chemotherapy [3]. Therefore intensive chemotherapy may have its greatest impact when used after clinical remission induced by conventional chemotherapy. Results of six studies, including our own, show that in patients with less than a complete response to conventional chemotherapy the rate of complete remission was 18 of 41 (44%) and did not differ from the reports in previously untreated patients. In patients who achieved complete remission by conventional chemotherapy the remission duration after high-dose chemotherapy was significantly longer than in patients with evidence of

Table 2. High-dose chemotherapy in SCC

Authors	Regimen	No. of patients treated	Status of disease	No. of CRs	Duration of CRs (months)
Spitzer *et al.* [16]	CTX 1.5 g/m² d 1–4 BCNU 300 mg/m² d 1 VP-16 125 mg/m² d 1–5	9	relapse	1	3
Pico *et al.* [18]	BCNU 300 mg/m² d 1 Melphalan 140 mg/m² d 5 Procarbazine 200 mg/m² d 1–4	5	relapse	2	NA
Phillips [19]	BCNU 1200 mg/m²	4	relapse	0	—
Spitzer *et al.* [20]	BCNU 300 mg/m² d 1 + 2	3	relapse	0	—
Douer *et al.* [21]	VBL 0.5 mg/m² d 1–2 CTX 1.5 g/m² d 3–4	1	relapse	1	2
Souhami *et al.* [22]	CTX 40–50 mg/kg d 1–4 XRT to primary	25	untreated	14	NA*
Farah *et al.* [23]	CTX 1.5 g/m² d 1–3 VP-16 200 mg/m² d 1–3 VCR 1.5 mg/m² d 1 + 3 ×2 ± ADR 80 mg/m² d 1	14	untreated	7	median 10†
Smith *et al.* [11]	7 g/m² d 1	27	intensification		
			15 < CR	4	median 2.5
			12 CR	—	median 8+
Klastersky *et al.* [24]	CDDP 120–180 mg/m² d 1	13	intensification		
	ADR 90–135 mg/m² d 1		8 < CR	6	median 4
	VP-16 240–360 mg/m² d 1–3		5 CR	—	median 10
Spitzer *et al.* [25]	CTX 1.5 g/m² d 1–3	10	intensification		
	VP-16 200 mg/m² d 1–3		6 < CR	3	median 6.5†
	VCT 1.5 mg/m² d 1 + 3		4 CR	—	median 10†
Stewart *et al.* [17]	CTX 120 mg/m²	10	intensification		
	TBI 800–1000 rad		5 < CR	2	4, 5
	± BCNU 150–600 mg/m²		5 CR	5	—
Ihde *et al.* [26]	XRT to primary (2000 rad/5 fractions)	8	intensification		
	CTX 60 mg/kg d 1–2		5 < CR	1	3
	VP-16 200 mg/m² d 1–3		3 CR	—	median 8
Present study	CTX 120 mg/m² over d 1 + 2	4	intensification		
	BCNU 400 mg/m² over d 1 + 2		4 < CR	2	3, 6
	VP-16 200 mg/m² d 1–5				

*Median survival in all responders: 6 months.
†Under maintenance chemotherapy.

residual disease prior to intensification [17, 24–26].

Based on our experience and that of others, high-dose chemotherapy with autologous marrow reinfusion has the potential to complement the effects of standard combination chemotherapy in SCC and is more likely to have its greatest impact when used as intensification in patients who have already achieved a complete remission.

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