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Short communication

A randomised phase II study of carmustine alone or in combination with tumour necrosis factor in patients with advanced melanoma

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Summary. Laboratory data suggest a synergistic interaction between carmustine (BCNU) and tumour necrosis factor (TNF) in melanoma. We therefore studied the activity of 200 mg/m^2 BCNU given alone or in combination with 88 μg/m^2 recombinant human TNF-alpha (rhTNFα) as a daily i.v. infusion for 5 days at 48-day intervals to patients with metastatic melanoma. In this randomised phase II trial, the rate of response to BCNU alone was 20% [95% confidence interval (CI), 2%–38%], and this was not improved by the addition of TNF (response rate, 10.5%; 95% CI, 1.3%–33%). Toxicity was higher in the combination arm, and there was no difference in survival.

Introduction

The results of systemic treatment for metastatic melanoma are disappointing, with overall response rates being low, complete remissions rare and response durations short. Dacarbazine, cisplatin, vindesine and nitrosoureas are the most active single agents, producing response rates of 15%–20% [1, 5, 13, 14]. Combination chemotherapy or high-dose intensive treatments are sometimes associated with significantly higher response rates of up to 45%, but without prolongation of survival [13, 14]. The results of treatment with biological agents are little better, with overall response rates to interferon-alpha or interleukin 2 ranging between 5% and 25%, depending on patient selection and schedule [6, 14, 15].

There is evidence in vitro and in vivo for interactions between some cytotoxic drugs such as Adriamycin, inhibitors of topoisomerase II and biological therapies, including tumour necrosis factor [2, 7]. Tumour necrosis factor-alpha (rhTNFα) is a 155 amino-acid peptide originally identified in the serum of mice that had been pre-treated with bacille Calmette-Guérin (BCG) and subsequently exposed to endotoxin [4]. Recombinant human TNFα is cytotoxic to many murine and human tumour cells in vitro, including some melanoma cell lines [8, 9]; however, the rates of response to TNFα have been disappointing, amounting to less than 1% in over 400 patients treated for a variety of tumours in phase I/II studies [10, 16].

We have shown in vitro and in animal studies that rhTNFα enhances the cytotoxicity of the nitrosourea carmustine (BCNU) against the murine melanoma B16 cell line and against human melanoma xenografts in nude mice [11]. This report describes the application of these observations to the clinical setting and presents the results of a randomised phase II study of rhTNFα given in combination with BCNU to patients with metastatic melanoma.

Patients and methods

Patients. Individuals aged 18–65 years were eligible for randomisation if they had histologically confirmed metastatic melanoma, progressive disease that was symptomatic or pre-symptomatic, a WHO performance status of 0–2 and a projected survival of >3 months. Patients with abnormal hepatic or renal function were included only if these abnormalities were due to the presence of metastatic disease and were not considered life-threatening. Subjects who had received prior chemotherapy were excluded from the study, as were those with a history of allergy because of the risks of allergic reactions to rhTNFα in atopic patients. The protocol was approved by the Ethics Committee of the Royal Marsden Hospital and all patients gave verbal, witnessed informed consent.

Treatment. BCNU (200 mg/m^2 in 250 ml saline) was given as an i.v. infusion over 1–2 h. The BCNU was made up by dissolving aliquots of 100 mg in 3 ml sterile ethanol and, after dissolution, adding 27 ml sterile water. The total dose to be given was diluted in 250 ml saline immediately prior to use. rhTNFα (PAC – 40) was provided by Asahi Chemical Industry Co. Ltd., Tokyo, as a sterile, dry preparation in vials containing 222 μg/vial. The contents of each vial were dissolved in 1 ml sterile water and further diluted in 100 ml normal saline immediately before use and were infused i.v. via a peripheral vein over 1 h. rhTNFα was given at a dose of 88 μg/m^2 daily for 5 consecutive days (days 1–5). This dose was selected on the basis of a phase I trial at the Royal Marsden Hospital in which 400 μg/m^2 was the maximum tolerated dose given as a single dose every 21 days [16]. Patients randomised to receive rhTNFα underwent an i.d. skin test with 500 units rhTNFα to test for hypersensitivity at 24 h before the first i.v. treatment, and those showing a positive

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Table 1. Patients' characteristics

	BCNU	BCNU+rhTNFα
Total number	20	19
Sex (M:F)	12:8	12:7
Median age (range)	47 years (27–68 years)	46 years (27–64 years)
WHO performance status:		
0	4	3
1	10	13
2	6	3
Sites of disease:		
Local	6	7
Lymph node	16	14
Skin	8	9
Pulmonary	9	8
Pleura	0	6
Liver	5	5
Bone	2	4
Adrenal	2	1
Other	7	3
Median time from diagnosis to entry in the study (range)	22 months (1–252 months)	25 months (1–156 months)
Median time from the first metastatic relapse to treatment (range)	3 months (0–156 months)	3 months (0–60 months)

Table 2. Response to BCNU and BCNU + rhTNFα

	BCNU	BCNU+rhTNFα
CR	1	1
PR	3	1
NC	0	1
PD	16	16
Overall response (CR+PR)	4 (16%)	2 (11%)
95% confidence limits	5 (36%)	1 (33%)

reaction were not to receive rhTNFα. Patients receiving the combination of BCNU and rhTNFα were given BCNU on day 3 (of 5) immediately after the third dose of rhTNFα.

All subjects received prophylactic antiemetic cover with metoclopramide prior to BCNU administration, but corticosteroids were not used because of the theoretical risk of their interfering with rhTNFα activity [3]. Patients receiving rhTNFα were also given 100 mg prophylactic ketoprofen p.o. at 1 h before rhTNFα treatment and 5–10 mg oral diazepam at 10 minutes prior to the rhTNFα infusion. Additional diazepam or pethidine was given as clinically indicated, e.g. in the event of rigors.

Treatment cycles were repeated at 6-week intervals for at least two courses. Responding patients and individuals with stable disease received a maximum of six cycles. Subjects showing clear evidence of progressive disease discontinued treatment at any stage.

Study design. Patients were randomised using a computer-generated, balanced randomisation programme with a block size of 4. Subjects were randomised centrally by telephone to the London Office of the Asahi Chemical Industry Co. Ltd.

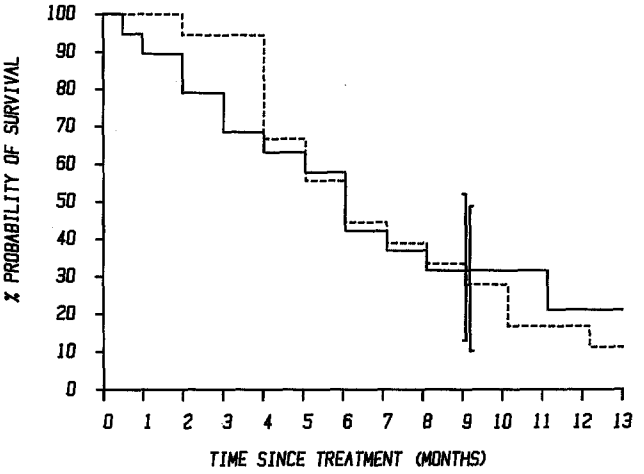

Fig. 1. Overall survival following treatment with BCNU (*solid line*) and BCNU + TNF (*dashed line*)

Assessment of response and toxicity. Patients were assessed prior to each treatment course by clinical examination, and quantitative measurements of lesions were made on alternate courses using the appropriate investigations [chest X-ray, ultrasound or computed tomographic (CT) scanning]. Subjects underwent a full blood count, urinalysis, liver-function tests, determinations of serum creatinine, urea and electrolytes as well as of [^{51}Cr]-ethylenediaminetetraacetic acid (EDTA) clearance and ECG prior to each course. The full blood count, serum creatinine, urea and electrolyte values and liver-function tests were in addition monitored weekly between courses. Toxicity was quantitated according to standard WHO criteria [17]. Patients receiving rhTNFα were monitored (pulse, blood pressure, temperature) throughout the administration of the drug, then hourly for 4 h and finally every 4 h for 24 h. Subjects receiving BCNU only were monitored every 4 h for 24 h on the day of treatment.

Response was defined according to WHO criteria [17] as follows: complete response (CR), the disappearance of all known disease as determined by two observations separated by no less than 4 weeks; partial remission (PR), a >50% decrease in the product of bidimensionally measured lesions as determined by two observations separated by no less than 4 weeks and the absence of new lesions; stable disease (SD), a <50% decrease and a <25% increase in the product of bidimensionally measured lesions; and progressive disease (PD), a >25% increase in the size of measurable lesions and/or the appearance of new lesions.

Statistical analysis. The chi-square test was used to assess differences between treatment groups, and Kaplan-Meier life tables were used to generate survival curves [12].

Results

A total of 41 patients were randomised, 20 to BCNU and 21 to BCNU + rhTNFα. As 2 individuals were randomised in error (both to the rhTNFα + BCNU arm), they did not receive treatment. Therefore, a total of 39 patients were evaluable for response and toxicity; however, both of the incorrectly randomised subjects were included in the survival analysis. Patients in the two arms were well matched for age, gender, performance status, sites of disease and time to entry into the study as shown in Table 1. None of the subjects had undergone prior chemotherapy. Patients received a median of 2 courses of BCNU (range, 1–5) and 2 courses of BCNU + rhTNFα (range, 1–6).

Four patients responded to BCNU alone [20%; 95% confidence interval (CI), 2%–38%; 1 CR, 3 PRs] and two

Table 3. Toxicity of BCNU and BCNU + rhTNFα expressed as the worst toxicity for each patient

Toxicity	BCNU WHO grade					BCNU + rhTNFα WHO grade				
	0	1	2	3	4	0	1	2	3	4
Haemoglobin	10	5	2	3	0	10	4	2	2	1
Leucopaenia	12	3	3	1	1	12	2	0	3	2
Lymphocytopaenia	18	0	1	0	1	15	1	2	1	0
Nausea/vomiting	6	7	7	0	0	6	0	10	3	0
Fever	19	0	1	0	0	14	4	1	0	0
Rigor	0	0	0	0	0	2	4	8	3	2
Hepatic	17	1	2	0	0	11	3	2	2	1
Respiratory	19	0	0	1[a]	0	16	2[b]	1[b]	0	0

[a] Acute dyspnoea due to pulmonary fibrosis
[b] Acute dyspnoea during rhTNF[a] infusion

responded to BCNU + rhTNFα (10.5%; 95% CI, 1.3%–33%; 1 CR 1 PR; Table 2); responses were seen in subcutaneous, pleural and pulmonary disease. This difference in response was not significant. The durations of response were 2, 5, 12 and 16+ months for BCNU alone and 5 and 15+ months for BCNU + TNF. There was no difference in median overall survival, which was 6 months for both treatment groups (Fig. 1).

The toxicity of both treatments is shown in Table 3. There was no difference in haematological toxicity or in the incidence of infection or emesis between the two arms. However, patients receiving BCNU + rhTNFα experienced the well-recognised acute toxicities that are associated with the infusion of TNFα, namely, fevers, rigors and hypotension. In addition, reversible elevations in hepatic transminases were seen.

Discussion

This is the first report of a randomised study to evaluate the effect of rhTNFα on a cytotoxic agent. We confirmed the activity of single-agent BCNU, recording a 26% overall response rate and 6 months' median survival from the start of treatment. These results are in close accord with the data published on the efficacy of single-agent nitrosoureas in melanoma [1]. The 6-month median survival for patients treated in the present study with single-agent BCNU at a standard dose of 200 mg/m^2 every 6 weeks is comparable with the 4-month median duration of survival achieved by nine patients previously treated at our institution with high-dose BCNU [13]. Subjects treated with high-dose BCNU therefore appeared to have no survival advantage over those treated with more standard schedules, despite the higher response rates (44%) [13].

Animal data in a murine melanoma model and in human melanoma xenografts have suggested that there is an interaction between BCNU and rhTNFα, resulting in a greater cytotoxic effect on tumours than that produced by either drug alone [11]. The present study shows that no such interaction exists in man when rhTNFα is given in this combination at a dose of 88 μg/m^2 daily for 5 days. The lack of activity seen in this study may be a consequence of rhTNFα dose used or its scheduling. The animal data would suggest that a higher dose of rhTNFα would be more effective, but phase I data have shown that such doses are prohibitively toxic to humans. A further explanation for the discrepancy between the results obtained in the animal model and the present findings is that in the former, tumours were always implanted at a site that may be more susceptible to systemic therapy, namely, a s.c. site. Visceral metastases, however, were predominant in this trial. It is also possible that the mechanism underlying rhTNFα-enhanced cytotoxicity is effective only in s.c. disease.

The use of randomisation in a phase II study has been valuable in this setting. Many trials of chemotherapy in melanoma involve the phase II methodology, with relatively small numbers of patients being entered. Confidence limits on response rates are often wide, and there can be bias in patient selection. Randomisation in phase II studies does not overcome the difficulties of wide confidence intervals but does reduce selection bias, therefore enabling better assessment of the treatment arm containing a novel therapy. A further advantage to randomisation in a phase II study is that it can be easily converted to a larger, more meaningful phase III investigation by a simple increase in the number of patients participating in the study.

There is currently great interest in combination cytokines, particularly rhTNFα, with chemotherapy. TNFα has been shown in vitro and in vivo to interact with a number of other cytotoxic drugs, including anthacyclines and alkylating agents, among others [2, 7]. We would caution the use of simple single-arm phase II methods in evaluations of novel cytokine-chemotherapy combinations.

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