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Granulocyte-macrophage colony-stimulating factor alone or with dacarbazine in metastatic melanoma: a randomized phase II trial

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Summary The potential antitumoural effect of granulocyte-macrophage colony-stimulating factor (GM-CSF) led us to evaluate GM-CSF alone or with dacarbazine (DTIC) in metastatic melanoma in first line randomized phase II. Treatment was arm A: GM-CSF: 5 μg kg^{-1}, bid, 14 consecutive days every 21 days and arm B: GM-CSF: 5 μg kg^{-1}, bid, day 2 to day 19 every 21 days and DTIC: 800 mg m^{-2}, day 1 of each cycle. 32 patients (pts) were included, 15 pts in arm A and 17 in arm B. All pts had visceral metastatic sites. 9 had only one metastatic site. The median number of cycles given was 2 in arm A and 3 in arm B. 100% and 89.4% of the planned dose of GM-CSF was given in arm A and arm B respectively. No objective response was obtained. 19 pts experienced at least WHO grade 3 toxicity. All pts had fever, 29 had a decrease in performance status and 23 had pain. Grade 3 toxicity were fever (38.7%), decrease in performance status (32.3%), pain (19.4%) and dyspnoea (12.5%). In this study, GM-CSF alone or in association with DTIC did not induce any antitumoural activity with subsequent toxicity. © 2001 Cancer Research Campaign http://www.bjcancer.com

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Granulocyte-macrophage colony-stimulating factor (GM-CSF) is a haematopoietic growth factor acting on early multipotent haematopoietic progenitors. GM-CSF regulates growth and differentiation to macrophages or neutrophils (Metcalf, 1985; Sieff, 1987). In prospective randomized trials, GM-CSF has shown a significant increase in neutrophil count and a significant reduction of neutropenia duration after chemotherapy or autologous bone marrow transplantation (Greenberg et al, 1996; Update of recommendations for the use of haematopoietic colony-stimulating factors: evidence-based clinical practice guidelines, 1996; Ravaud et al, 1998). Moreover, GM-CSF induces mobilization and facilitates collection of peripheral blood stem cells (Demirer et al, 1996).

Furthermore, there is some evidence that GM-CSF could have antitumoural activity by enhancing immune response. In vitro GM-CSF activates monocytes and macrophages to be cytotoxic to human cell lines (Grabstein et al, 1986; Mazumder et al, 1990; Williams et al, 1994). GM-CSF induces antibody-dependent cell-mediated cytotoxicity (Kushner and Cheung, 1989; Mazumder et al, 1990; Nagler et al, 1994). Proliferation of interleukin-2 driven T lymphocytes is enhanced in the presence of GM-CSF (Santoli et al, 1988).

In experimental animal models, GM-CSF has been found to enhance the cytotoxicity of macrophages through production of nitric oxide and free radicals (Hill et al, 1993). Moreover, gene transfection of GM-CSF into tumour cells induces an antitumour activity and protection of mice when challenged with tumours (Dranoff et al, 1993). In humans, GM-CSF enhances cytotoxicity of T lymphocytes CD3+ CD16- following bone marrow transplantation (Richard et al, 1993). Antibody-dependent cell-mediated cytotoxicity is also increased (Ragnhammar et al, 1993). The ability to stimulate presentation of antigens is supported by GM-CSF enhancing MHC class II expression on monocytes (Willman et al, 1989). Moreover, GM-CSF is one of the main cytokines used for producing ex vivo dendritic cells, which act as professional antigen-presenting cells to induce T cell immune response (Szabolcs et al, 1995; Bender et al, 1996; Coulon et al, 2000). When GM-CSF was used during antitumoural vaccination with peptide in patients with melanoma, it enhanced the immune response through activation of CD4 + and CD8 + T cells (Jager et al, 1996).

The efficacy of GM-CSF as an antitumour agent has been hypothesized in a prospective trial in patients with soft-tissue sarcomas treated with chemotherapy in association with GM-CSF for prevention of haematological toxicity. One study showed the unexpected frequency of partial and complete response that could account for an antitumoural effect of molgrastim (Edmonson et al, 1994, 1997).

Prognosis of metastatic melanoma is poor and median survival is 6–8 months (Balch et al, 1997). Palliative treatment is based on chemotherapy with dacarbazine as first-line treatment (Ho, 1995). The arguments suggesting the efficacy of non-specific immunotherapy in melanoma are that interleukin 2 has been shown to induce clinical responses and long durable responses in

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metastatic melanoma (Legha et al, 1998; Atkins et al, 1999) and that interferon alpha induces a significant increase of disease-free survival and a possible increase in survival when used as adjuvant treatment (Kirkwood et al, 1996).

Due to a possible antitumoural effect of GM-CSF through an immune modulation and to low efficacy of chemotherapy in metastatic melanoma, we conducted a multicenter randomized trial comparing GM-CSF alone and GM-CSF associated with dacarbazine in metastatic melanoma patients.

PATIENTS AND METHODS

Patients

Adult patients under 70 years of age were eligible if they had histologically confirmed melanoma with distant metastatic disease that could be measured in 2 dimensions. Patients had an Eastern Cooperative Oncology Group (ECOG) performance status ≤ 2. No prior chemotherapy was allowed. Previous adjuvant interferon alpha was allowed, but a delay of more than 6 months was required before inclusion. Patients with central nervous system metastases were excluded, except if they had been previously treated (surgery and/or radiotherapy) and subsequently demonstrated stable disease or no evidence of recurrence. Biological inclusion criteria were leukocyte count $\geq 4 \times 10^9$ l^{-1}, haematocrit level ≥ 30%, platelet count $\geq 100 \times 10^9$ l^{-1}, creatinine concentration < 150 μmol l^{-1} and transaminases < 2-fold the upper limit of normal values except in the case of hepatic metastases where < 5 fold the upper limit of normal values was required. Furthermore, patients with severe infection or positivity for HIV or hepatitis B or C were not eligible. Glucocorticosteroids were not allowed. Patients did not have any additional malignancy other than cervical carcinoma in situ or basal cell carcinoma. Pregnant or lactating women were excluded. Patients had to give written informed consent before entry into the trial.

Pretreatment evaluation

Clinical history and physical examination were recorded for all patients. Before inclusion, patients had a staging including thoracic and abdominal computed tomographic scans and a chest X-ray.

Study design and statistical considerations

The study was an unblinded randomized phase II trial conducted in 3 centres with no stratification. Registration and randomization of eligible patients were performed at the data monitoring centre of Institut Bergonié. The primary end point was the objective response rate, and secondary end points were response duration, toxicity and the response with dacarbazine alone after failure with GM-CSF alone.

An objective response rate of at least 20% would be of sufficient interest to encourage further investigation of GM-CSF. The determination of the numbers of patients followed Gehan's rules, which indicates that including 14 patients gives a 95% chance of detecting at least one response if the response rate was ≥ 20% (Gehan, 1961). If at least one response occurred in the first 14 patients, it was planned to increase the number of patients up to a total of 25 patients per arm to assess the response rate more precisely.

Treatment

Eligible patients were randomly assigned to receive either GM-CSF or GM-CSF with decarbazine (GM-CSF + DTIC). Dosage and schedule of GM-CSF were based on the trial of Edmonson et al (1994, 1997). Patients in the GM-CSF arm received subcutaneous GM-CSF at a dose of 5 μg kg^{-1}, bid for 14 consecutive days every 21 days. Patients in the GM-CSF + DTIC arm received DTIC at a dose of 800 mg m^{-2} on day one and subcutaneous GM-CSF at a dose of 5 μg kg^{-1}, bid from day 2 to day 19 every 21 days.

Response was assessed every 2 cycles. Work-up was identical to the initial pretreatment evaluation. Each time stabilization or an objective response occurred, 2 additional cycles of treatment were planned up to a total of 12 cycles.

Patients who progressed under GM-CSF alone received DTIC at a dose of 800 mg m^{-2} on day one every 21 days.

Evaluation of treatment

The World Health Organization (WHO) criteria were used to determine tumour response (Miller et al, 1981). Complete response (CR) was defined as the complete disappearance of all measurable and evaluable tumour sites for at least 4 weeks. The duration of CR was calculated from the first date of documentation of CR to the date of the first evaluation of disease progression. Partial response (PR) was considered to be a ≥ 50% decrease in the sum of products of the greatest perpendicular diameters that lasted for at least 4 weeks, with no increase in known lesions and without appearance of any new lesions. The duration of PR was calculated from the first day of treatment. When the evaluation showed a decrease in lesions < 50% and an increase < 25%, patients were considered to have a stable disease (SD). Progressive disease (PD) was considered to be when any lesion increased by ≥ 25% or when a new lesion appeared. Patients who presented with a CR, PR or SD were further evaluated every 2 to 3 months during the first year and then every 4 to 6 months. Survival duration was evaluated from the start of treatment to the date of the last contact or the date of death. Toxicities encountered were classified according to the World Health Organization grading system.

RESULTS

Patient characteristics

32 patients were entered into this study (Table 1). All were evaluable for toxicity. 2 were considered not to be assessable for tumour response: one had a grade 3 decrease in general status and went off the study, and another refused to continue due to grade 3 pain after one cycle of treatment.

Administration of treatment and toxicity

During treatment, the median dose of GM-CSF given in the GM-CSF arm was 8960 μg (range: 3300–11 200) corresponding to 100% of the planned dose, while the median dose of GM-CSF in the GM-CSF + DTIC arm was 12 070 μg (range: 7820–17 340), accounting for 89.4% of the planned dose. The median number of cycles received was 2 in the GM-CSF arm and 3 in the GM-CSF + DTIC arm. 3 patients (20%) in the GM-CSF arm and 11 (64.7%) in the GM-CSF + DTIC arm had a dose modification of GM-CSF due to the induced toxicity. 6 patients (40%) in the GM-CSF arm

Table 1 Patient characteristics

Characteristics	GM-CSF arm	GM-CSF + DTIC arm
No. of patients	15	17
Sex (male/female)	11/4	11/6
Age, y. Median (range)	56 (35.5/70.5)	57 (36.6/67.6)
Site of melanoma		
Cutaneous	11	12
Ocular	1	2
Visceral	1	0
Unknown	2	3
Time from diagnosis to metastasis Median (range) months	17.5 (0/89)	17.9 (0/140.5)
ECOG performance status		
0	7	7
1	7	8
2	1	2
Pts with visceral metastatic sites	15	17
Sites of metastatic disease		
Lung	7	13
Superficial lymph nodes	1	2
Deep lymph nodes	6	9
Liver	5	7
Bone	2	5
Others	9	4
No of metastatic sites		
1	5	4
2	6	6
> 2	4	7

while 13 (76.5%) in the GM-CSF + DTIC arm had a WHO grade 3 or 4 toxicity (Table 2).

Fever (32 patients), decrease in performance status (29 patients) and pain (23 patients) were the most common adverse events related to GM-CSF in the 2 groups. Only 3 patients had a grade 4 toxicity with anaemia (2 pts) and vomiting (1 pt), while 6 patients in the GM-CSF arm and 10 in the GM-CSF + DTIC arm had a grade 3 toxicity. The grade 3 toxicities were fever (6 patients in both groups), pain (6 patients in the GM-CSF + DTIC arm; 1 in the GM-CSF arm), decrease in performance status (7 patients in the GM-CSF + DTIC arm; 3 in the GM-CSF arm), dyspnoea (4 patients in the GM-CSF arm) and thrombocytopenia (2 patients in the GM-CSF + DTIC arm).

Response to treatment and survival

15 were included in the GM-CSF arm while 17 were included in the GM-CSF + DTIC arm. Among 30 assessable patients, no patient achieved an objective response. The best response achieved was stable disease in 3 patients treated with GM-CSF + DTIC, while all other patients already had tumour progression at the evaluation done after 2 cycles of treatment.

Among 8 patients who received DTIC alone after failure with GM-CSF alone, one patient had a partial response.

The median follow-up was 44.2 months. The median overall survival was 6.3 months; 6.3 months in the GM-CSF arm and 7.3 months in the GM-CSF + DTIC arm.

DISCUSSION

In this study, GM-CSF induced no clinically evaluable antitumoural activity in patients with metastatic melanoma, given either alone or in association with standard chemotherapy by dacarbazine. This is the first study reporting the clinical evaluation of GM-CSF alone or in association with chemotherapy in metastatic melanoma patients.

Table 2 Grade 3–4 WHO toxicity

	GM-CSF		GM-CSF + DTIC	
Toxicity	Gr 3	Gr 4	Gr 3	Gr 4
Fever	6	0	6	0
Pain	1	0	6	0
Decrease in performance status	3	0	7	0
Dyspnea	0	0	4	0
Skin disorders	0	0	0	0
Nausea / vomiting	0	0	0	1
Hypotension	0	0	0	0
Neurological	3	0	0	0
Anaemia	0	0	1	2
Thrombocytopenia	0	0	2	0
Hypercreatininaemia	0	0	0	0

Other authors have shown a dramatic increase in response rate with subcutaneous GM-CSF associated with chemotherapy (ifosfamide, cisplatinum and doxorubicin) in patients with sarcoma (Edmonson et al, 1994, 1997). However our study does not confirm that GM-CSF increases the efficacy of a given chemotherapy in metastatic solid tumours.

In the phase I trial reported by Edmonson, 15 patients were treated with chemotherapy supposed to induce 30% objective response and complete response less than 5%. 14 patients had metastatic localization. An objective response was seen in 7 patients. Furthermore, 5 patients had a complete remission and another had a complete remission on metastatic site but not on the primary tumour. The median duration of survival was 27 months, longer than expected. Another phase II study from the EORTC showed in 104 patients with metastatic sarcoma, that GM-CSF added to escalated doses of chemotherapy could increase the expected response rate to 45% and 10% of complete response (Steward et al 1993); nevertheless, the impact of GM-CSF as well as the dose intensity of chemotherapy are points to be debated. Except for sarcomas, where the chemotherapy used is highly effective (objective response $\geq 30\%$), the wide literature so far on the topic of GM-CSF given to prevent haemotoxicity, has not provided data suggesting tumour efficacy of GM-CSF in metastatic solid tumours.

Our findings do not exclude the possibility of GM-CSF having antitumoural activity on a lower tumour burden or in association with a treatment devoted to triggering an immune response. This hypothesis has since received confirmatory data on the ability of GM-CSF to enhance immune response, even though there is not yet a major impact on clinical outcome. The main focus of GM-CSF activity on immune response has concerned the impact of GM-CSF on the recruitment, activation and survival of dendritic cells. It is known that GM-CSF given by intradermal route, leads to local skin reactions, with a progressive accumulation of Langerhans cells, while the subcutaneous route failed to do so (Kaplan et al, 1992). It has been shown that GM-CSF enhances the recruitment of dendritic cells, thereby increasing the number of dendritic cells in regional lymph nodes, providing contact with lymphocytes (Kass et al, 2001). In addition, GM-CSF enhances the activation and survival of dendritic cells (Armitage, 1998). Moreover, GM-CSF protects dendritic cells from apoptotic death (Rabinovich et al, 1999).

The induction of an immune response by GM-CSF given intradermally can be detected in eliciting local skin reaction and strong DTH response, when used alone (Kaplan et al, 1992) or as adjuvant therapy with peptide (Jäger et al, 1996; McNeel et al, 1999), a finding already demonstrated in experimental models (Disis et al, 1996).

Furthermore, there are arguments for stimulation of T lymphocytes. When the GM-CSF was given intralesionally in subcutaneous metastases, an increase in T cells, especially CD4+ cells was noticed at the tumour site (Miller et al, 1981). Added to vaccination procedures, either to peptide (Jäger et al, 1996) or to irradiated autologous tumour cells (Si et al, 1996), GM-CSF induced an increase in the CTL response.

The increase in immune response, as shown biologically, has been associated in clinical trials with tumour response.

It has been shown that GM-CSF given intralesionally in subcutaneous metastases induced 3 partial responses in 13 patients with metastatic melanoma (Miller et al, 1981). GM-CSF associated with vaccination either with peptide or with irradiated autologous tumour cells has shown to increase the clinical response to peptide alone (Jager et al, 1996) or to irradiated autologous tumour cells with BCG (Si et al, 1996). GM-CSF given intradermally with peptides derived from Melan A/MART-1, tyrosinase and gp 100 in HLA-A2+ patients with metastatic melanoma induced one complete response and 2 partial responses in 3 patients, while vaccination with the same peptide alone induced only stabilization. Furthermore, in 20 patients with metastatic melanoma treated by vaccination with autologous irradiated tumour cells associated with BCG and GM-CSF, 2 patients had a complete response and 2 others a partial response, while a prior study by vaccination with autologous irradiated tumour cells associated with BCG did not show any clinical response (Leong et al, 1999). In the latter study (Leong et al, 1999), GM-CSF added to a treatment designed to trigger an immune response, was effective for large metastatic tumour burden from melanoma.

On the other hand, another study demonstrated for efficacy with GM-CSF in a lower tumour burden, while given as adjuvant to surgery stage III or IV melanoma patients, but the results have to be taken with caution (Spitler et al, 2000). 48 patients with stage III or IV melanoma rendered disease-free by surgery received GM-CSF in an adjuvant setting. Compared with matched historical controls, overall survival and disease-free survival were significantly prolonged in patients receiving GM-CSF. The major concern of the Spitler study is that it is an uncontrolled trial, even compared to matched historical controls. The metastatic patients in it were from a very selected population, rendered macroscopically disease-free by surgery, which is far from the general population, especially patients included in our study. No conclusion can be drawn out from the study, except that immunobiologic modifier drugs are probably more active in low tumour burden.

The toxicity induced by the dosage and schedule in this study was similar to that reported in the initial sarcoma study with identical dosage and schedule (Edmonson et al, 1994, 1997). In both arms of our study, fever, decrease in performance status and pain were the most frequent grade 3 side effects for patients accounting for 38.7%, 32.3% and 19.4% respectively. In the initial report (Edmonson et al, 1994), toxicities were reported without grading of severity. Nevertheless, 10 of the 15 patients had to receive prednisone to allow continuation of treatment.

CONCLUSION

In this study of patients treated for metastatic melanoma, GM-CSF alone or with dacarbazine, at the dosage and schedule used, did not induce any objective response. Toxicity was severe with 19 patients (59.3%) presenting at least a WHO grade 3 toxicity. Therefore, these findings do not support the contention that GM-CSF has an antitumoural activity in metastatic solid tumours, at least where there is a large tumour burden.

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