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# A Comparison of Two GM-CSF Schedules to Counteract the Granulo-monocytopenia of Carboplatin–Etoposide Chemotherapy

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**In order to obtain the beneficial effects from granulocyte–macrophage colony-stimulating factor (GM-CSF) on granulo-monocyte recovery with the minimum dose and toxicity, we compared the effect of two different GM-CSF schedules (5 μg/kg/day subcutaneously, days 5 to > 18 versus days 12 to > 18 on the cytopenias which follow cytostatic treatment with carboplatin (400 mg/m² intravenous (i.v.) day 1) and etoposide (100 mg/m² i.v. days 1 to > 3). 13 patients entered the study for a total of 36 evaluable cycles. The cytostatic treatment produced a neutropenia that persisted for up to day 22 (absolute neutrophil count (ANC) < 1000/μl in 25% and ANC < 2000 in 50% of control cycles). Early GM-CSF administration markedly increased the leucocyte nadir and produced two waves of leucocytosis: an early one, linked to marrow reserve release and presumably of no value to the patients; and a delayed one, due to marrow precursor and progenitor cell proliferation, in which the granulo-monocytosis was associated with a marked eosinophilia. The delayed GM-CSF administration markedly increased the leucocyte nadir and accelerated granulo-monocyte recovery (with an only modest eosinophilia), so that chemotherapy could be repeated every 21 days in all the patients.**

**Key words: GM-CSF, cancer chemotherapy, neutropenia, dose intensity, carboplatin, etoposide**
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## INTRODUCTION

GRANULOCYTE–MACROPHAGE colony-stimulating factor (GM-CSF) stimulates the proliferation and differentiation of haematopoietic cells with an action that extends from the progenitors to precursors and mature cells of the granulo-monocyte lineage [1–5]. Its administration allows the severity and duration of granulo-monocytopenia, which follows cytostatic treatment, to be notably reduced [6, 7].

Generally, the administration of GM-CSF (or other growth factors) is started shortly after the end of cytostatic therapy, and

continued until the leucocyte count has been normalised. This approach is reasonable following high-dose cytostatic treatment (with or without marrow transplantation), where the risk of infection is high and cost and toxicity problems are negligible [7–11].

During repeated cycles of standard or moderately high-dose chemotherapy, the administration of GM-CSF or of other haemopoietic growth factors aims at accelerating the recovery of the leucocyte count in order to allow the preset dose intensity to be maintained [12–16]. Under these circumstances, an accurate scheduling of GM-CSF is fundamental in order to obtain the desired effect with the minimum possible dose.

With this objective, we studied two different GM-CSF schedules in patients treated with carboplatin and etoposide, comparing the effect of treatment that was begun immediately after chemotherapy with that of treatment begun shortly before the leucocyte nadir. The aim of the study was to determine whether the short GM-CSF schedule was sufficient to accelerate the recovery of neutrophil counts so that chemotherapy could be restarted by day 22.

## PATIENTS AND METHODS

### *GM-CSF*

Human recombinant *E. coli*-derived GM-CSF in the non-glycosylate form, purified and freeze-dried, was supplied by Sandoz/Schering-Plough (Basel, Switzerland/Kenilworth, New Jersey, U.S.A.).

### *Study programme*

13 patients with solid tumours (Table 1), were randomised with a cross-over design. The sample size allowed the demonstration of detectable difference delta/sigma = 1, with minimum correlation of 0.5, a power of 80% and an $\alpha$ error of 0.05 [17]. The criteria for inclusion in this study were as follows: histological diagnosis of solid tumour, age between 18 and 70 years, performance status 0–1 (ECOG), normal liver and kidney function, adequate bone marrow reserve (white blood cells (WBC) > 4000/μl, with absolute neutrophil count (ANC) > 2000, platelets > 120 000/μl), normal cardiac function and signed informed consent. As well as objective examinations and routine laboratory research carried out before each therapy cycle, the patients underwent specific examinations in relation to the type of pathology presented.

A blood cell count was carried out on days 1, 6, 8, 11, 13, 15, 18, 22 and 29 of each cycle. The patients received three cycles of chemotherapy according to the following programme: carboplatin 400 mg/m² intravenously (i.v.) day 1 and etoposide 100 mg/m² i.v. days, 1, 2, 3. Each cycle was repeated every 28 days, in the presence of permissive haematological parameters (WBC > 4000/μl, platelets > 120 000/μl).

*Table 1. Patients' characteristics*

| | |
|---|---|
| No. patients included | 13 |
| Mean age (years) | 58 (range 20–68) |
| Male/Female | 11/2 |
| Previous radiotherapy | 2 |
| Previous chemotherapy | 0 |

In two of the three cycles, recombinant human GM-CSF (rhGM-CSF) 5 μg/kg/day subcutaneous (s.c.) was added from day 5 to 18 or from day 12 to 18. The study was open-label and a simple randomisation list was applied. Table 2 shows the sequence of treatment for each group.

*Table 2. Study design*

| Patients | Cycle sequence |
|---|---|
| 1–7 | A B C |
| 2–8 | B C A |
| 3–9*, 13 | C B A |
| 4–10 | A C B |
| 5–11 | B A C |
| 6–12 | C A B |

Cycle A: CBDCA + VP16 + rhGM-CSF days 12–18;
Cycle B: CBDCA + VP16 + rhGM-CSF days 5–18;
Cycle C: CBDCA + VP16.
*Patient withdrawn for toxicity.

### *Statistical evaluation*

The data were analysed with SYSTAT 5.0 package (DOS version) by means of the MGLH procedure [18]. The model takes into account both period and carry-over effect [19]. The contrasts were realised by means of the Scheffè method.

### *Toxicity*

The adverse events linked with rhGM-CSF were evaluated according to the WHO criteria [20]. The therapeutic response and the toxicity linked to the cytostatic treatment are not shown as they do not represent the primary end-point of the study.

## RESULTS

Figure 1 shows the effect of the chemotherapy ± GM-CSF on various haematological parameters.

In the control group, chemotherapy produced a leucothrombocytopenia with the nadir on day 15 (Figure 1). In 42% of the cycles, the neutrophil nadir was less than 1000/μl and in 16% less than 500/μl. Recovery was relatively slow. On day 22, in 50% of the cycles, the neutrophil count was less than 2000/μl (< 1000 in 25%).

With the administration of GM-CSF from day 12, the neutrophil nadir (with only 8% of the cycles with ANC < 1000) was significantly increased compared to controls ($P < 0.05$, days 13, 15) and leucocyte recovery was accelerated: on day 22 all patients had an ANC > 2000 (Figure 1a, Table 3). This accelerated leucocyte recovery involved not only the neutrophils but also the monocytes and the eosinophils (Figure 1b, c).

Early administration (from day 5) of GM-CSF modified the entire pattern of postchemotherapy WBC: the curve became biphasic. Early leucocytosis was followed by a reduction in the leucocyte count with a very high nadir on day 15 and a successive second phase of leucocytosis. This was sustained by a neutrophilia associated with an important eosinophilia and a moderate monocytosis (Figure 1a–c).

The three study groups were not noticeably different for haemoglobin, reticulocyte, lymphocyte (not shown) and thrombocyte values (Figure 1d).

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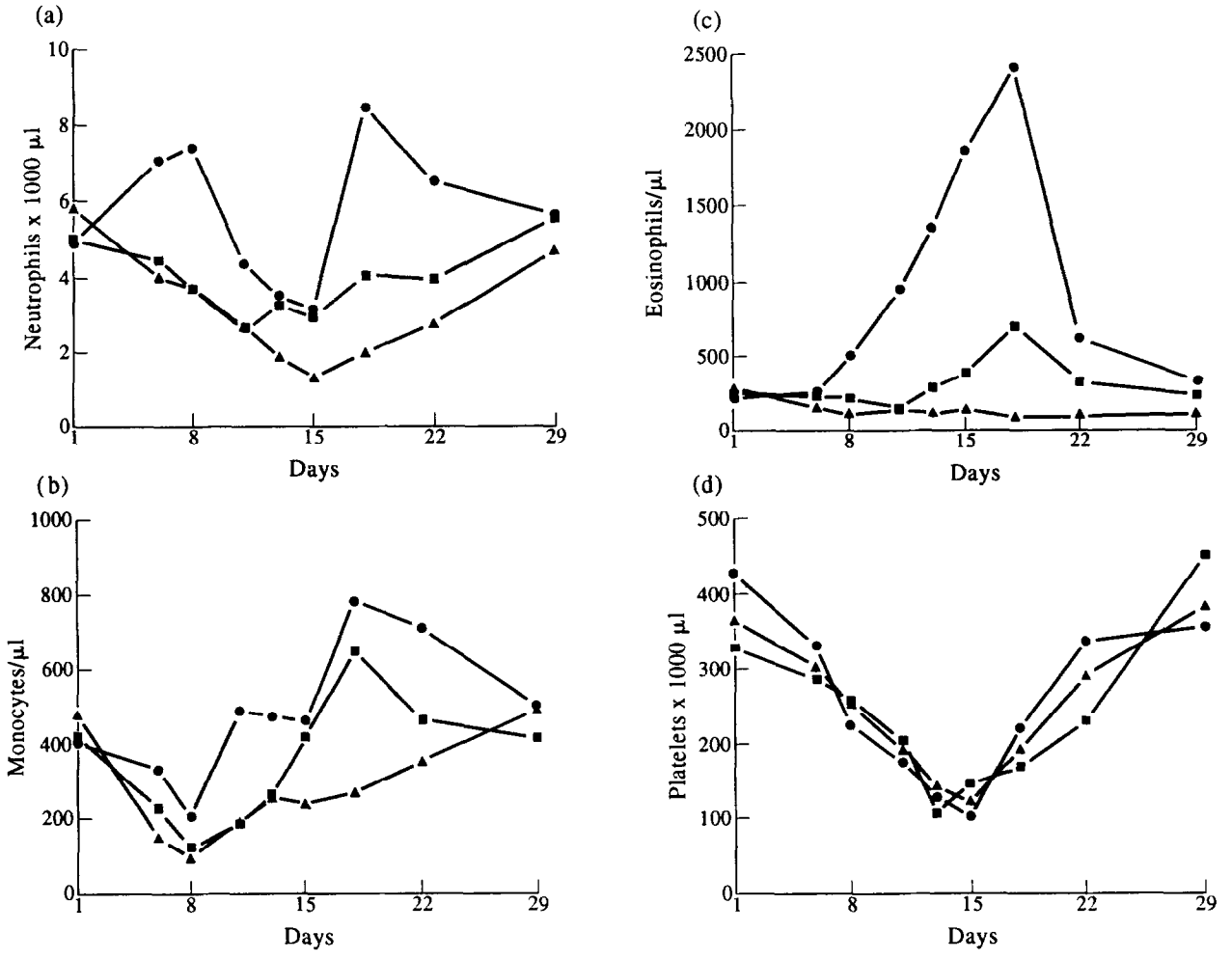


**Figure 1. Effect of chemotherapy ± GM-CSF on the main haematological parameters of the patients studied (means of 12 values per point). (a) Neutrophil count; (b) monocyte count; (c) eosinophil count; (d) platelet count. -▲-, control cycles; -■-, GM-CSF days 12–18; -●- GM-CSF days 5–18.**

Table 4 shows the principle toxicities linked to the treatment. In 1 patient, GM-CSF had to be suspended at the third administration because of allergy. Treatment was reasonably well tolerated by the other patients, with level 2–3 toxicity only in cycles when GM-CSF was administered from days 5 to 18.

## DISCUSSION

Chemotherapy with carboplatin and etoposide is widely used in oncology. Its major limitation is haemopoetic toxicity [21, 22]. Even with the moderate doses that we used in patients with a good performance status, it produced a neutropenia that persisted even at day 22 postchemotherapy (ANC < 1000 in 25% and ANC < 2000 in 50% of the control cycles).

GM-CSF, because of its stimulating action on the proliferation and differentiation of haematopoietic progenitors, and on the release into the circulation of bone marrow neutrophils, counterbalanced the granulo-monocytopenia of the cytostatic therapy. With administration starting immediately after chemotherapy, the leucocyte nadir was prevented and two leucocytosis curves were noted: the first is probably due to marrow reserve release, whereas the second is the result of the stimulation of bone marrow cell proliferation.

Delaying GM-CSF treatment produced a more modest effect,

*Table 3. Absolute neutrophil counts (ANC) at different days of treatment (values/µl ± S.D.)*

| | Days of treatment | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | 1 | 6 | 8 | 11 | 13 | 15 | 18 | 22 | 29 |
| Control | 5819 ± 2766 | 4007 ± 2110 | 3723 ± 2471 | 2721 ± 1880 | 1909 ± 1202 | 1356 ± 953 | 2034 ± 2045 | 2821 ± 2114 | 4710 ± 2431 |
| GM-CSF days 12–18 | 4995 ± 1450 | 4446 ± 2088 | 3681 ± 1743 | 2624 ± 1252 | 3269 ± 1500 (*) | 2958 ± 1957 (*) | 4060 ± 4434 | 3972 ± 2114 | 5541 ± 3197 |
| GM-CSF days 5–18 | 4873 ± 3077 | 7070 ± 3679 (*†) | 7407 ± 5222 (*†) | 4383 ± 3079 (*†) | 3521 ± 1564 (*) | 3178 ± 1830 (*) | 8490 ± 5807 (*†) | 6532 ± 3347 (*†) | 5669 ± 2859 |

*$P < 0.05$ compared to controls; †$P < 0.05$ compared to treatment from day 12 to 18.
$P$ values were calculated with the Scheffè method (see statistical evaluation).

*Table 4. Toxicities*

| | | WHO Toxicity grades | | | |
|---|---|---|---|---|---|
| | 0 | 1 | 2 | 3 | 4 |
| Skin | 8 | 1 (8%) | 4 (31%) | | |
| Allergic reaction | 12 | – | – | 1 (8%) | – |
| Fever | 9 | 4 (31%) | – | – | – |
| Fatigue | 11 | 1 (8%) | – | 1 (8%) | – |
| Myalgias/arthralgias | 11 | 2 (15%) | – | – | – |
| Nausea | 10 | 3 (23%) | – | – | – |
| Lack of appetite | 11 | 2 (15%) | – | – | – |
| Pain (headache) | 10 | 3 (23%) | – | – | – |
| Hypotension | 10 | – | 2 (15%) | 1 (8%) | – |

in fact, the neutrophil counts were significantly different in the two groups of GM-CSF-treated patients in various treatment phases ($P < 0.05$ days 6, 8, 11, 18, 22).

However, if a more detailed comparison is made between these two GM-CSF schedules, it can be observed that early administration produced an early neutrophilia that is presumably of no value to the patients; and a marked eosinophilia, the clinical significance of which is not known, although the pathological pattern of hypereosinophil syndromes should be kept in mind.

Delayed administration of GM-CSF, in a period when there has already been a partial reconstitution of the progenitor and precursor bone marrow pool, almost totally prevents the leucocyte nadir, and accelerates the neutrophil recovery in a manner that could allow chemotherapy to be resumed in all the patients on day 22 (using both 1000 and 2000 ANC as endpoints).

With regard to monocytopenia, the difference between the two GM-CSF schedules was modest and mainly concerned the first postchemotherapy days.

Therefore, a brief course of treatment with GM-CSF, beginning immediately before the nadir when the bone marrow contains an elevated number of committed progenitors and precursors, i.e. the main targets of GM-CSF [23], is capable of producing a haemopoietic stimulus that is more than sufficient to counterbalance the myelotoxicity of the traditional chemotherapy treatment that we studied, such as carboplatin and etoposide. This abbreviated schedule, with undoubted advantages of reduced costs, still should allow a correct dose intensity to be administered. Moreover, on a larger scale, reduced toxicity from GM-CSF can be anticipated. It is likely that the best GM-CSF schedule depends on the type of chemotherapy used. Therefore, the use of this short GM-CSF schedule in association with different chemotherapy programmes deserves further investigation, to address the issue of cumulative haemopoietic toxicity.

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