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SHORT REPORT

Unexpected remission of acute myeloid leukaemia after GM-CSF

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Summary. The administration of granulocyte-monocyte colony-stimulating factor (GM-CSF) was associated with complete clinical and haematological response in an adult patient with minimally differentiated acute myeloid leukaemia who presented with pneumonia and moderate neutropenia, but no blast cells in the peripheral blood. The response lasted 9 months. At relapse, a second GM-CSF course resulted in a very good partial remission lasting 5 months, although differences in the kinetics of haemoglobin, neutrophil and platelet recovery were noted. Subsequent recurrences were managed with chemotherapy, a complete remission being obtained twice more and lastly consolidated with myeloablative chemo-radiotherapy supported by a peripheral blood stem cell autograft. This report suggests that GM-CSF should be further investigated as a therapeutic agent in selected cases of AML.

Keywords: GM-CSF, AML, pneumonia, remission.

Granulocyte-macrophage colony-stimulating factor (GM-CSF) is one of several cytokines involved in the control of normal haemopoiesis, acting as powerful promoter of myeloid cell proliferation and differentiation (Sieff *et al*, 1985). The therapeutic value of recombinant human GM-CSF has been assessed in a variety of neutropenic disorders predisposing to infections, including granulocytopenia caused by cancer chemotherapy or secondary to clonal haematological disorders such as acute myeloid leukaemia (AML) and primary myelodysplastic syndromes (Cannistra *et al*, 1989; Ganser *et al*, 1989; Estey *et al*, 1991, 1992). In AML, GM-CSF appears to stimulate the growth of clonogenic cells *in vitro* (Vellenga *et al*, 1987; Preisler *et al*, 1993), and may promote disease acceleration *in vivo*, thereby reducing the likelihood of response to chemotherapy (Estey *et al*, 1992). Presently, use of GM-CSF cannot be recommended as a first therapeutic step in AML. In contrast, a recent report showed that patients with myeloid leukaemias relapsing after an allogeneic bone marrow transplantation and treated with G(granulocyte)-CSF only, can achieve a complete response with restoration of haemopoiesis of donor origin (Giralt *et al*, 1993). This observation raises the important issue of a direct therapeutic use of G-CSF in AML. We observed an adult patient with minimally differentiated AML in whom a complete clinical and haematological response of worthwhile duration was unexpectedly achieved at presentation and first relapse with GM-CSF only.

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CASE REPORT

A previously healthy 52-year-old woman became pyrexial in October 1990. A chest film disclosed extensive pneumonia involving the left lower lobe. Total white blood cell count was 1×10^9/l with $0{\cdot}76 \times 10^9$/l granulocytes and no circulating blast cells. Examination of bone marrow revealed an almost total replacement of normal cellularity by undifferentiated agranular blast cells. A trephine marrow biopsy showed 90% infiltration with blast cells of non-lymphoid type, very little residual normal haemopoiesis, no increase in reticulin fibres, and no dysplastic changes. Blasts were myeloperoxidase (MPO) and alpha-naphtyl butyrate esterase unstained. They could not be obtained in sufficient numbers for an immunophenotypic analysis. The cytogenetic study gave no analysable metaphases. At first relapse the immunophenotype of bone marrow blasts was reassessed on a FACScan analyser (Becton Dickinson, Mountain View, Calif.), using a panel of monoclonal antibodies for acute leukaemia characterization. Although the analysis was compounded by contaminating peripheral blood mature T-lymphocytes (CD3 65%), interestingly it showed a discrepancy between CD13 (20%) and CD33 (4%) antigen-positive cells. This peculiar pattern was confirmed at second relapse, when blast cells present in the peripheral blood were confirmed to express only CD13 (40%) and not CD33 (1%) antigens, the CD34 (48%) stem cell antigen and HLA-DR (67%). A diagnosis of MPO-AML was eventually made, with the infrequent $CD13^+$ and $CD33^-$ surface immunophenotype as detected in 2/10 M0 (minimally differentiated) AML

cases reported by the French–American–British group (Bennett *et al*, 1991).

At presentation, the administration of cytotoxic chemotherapy was felt inappropriate in this patient with extensive pneumonia. A broad-spectrum antibiotic combination was started. 1 week later the patient was apyretic with partial clearing of her pneumonia. Blood counts were unmodified, with total neutrophils $0{\cdot}73 \times 10^9$/l and no circulating blast cells. Because AML patients presenting with pneumonia have a significantly reduced probability of survival (Estey *et al*, 1982), treatment with GM-CSF was considered in this patient without blast cells detectable in the peripheral blood and whose moderate neutropenia indicated the existence of normal residual granulopoiesis. It was assumed that GM-CSF would induce both numerical increase and activation of neutrophils; this in turn leading to further clinical improvement. To cope with a potential stimulation of AML, GM-CSF would be stopped when $MPO^-/CD13^+/CD33^-$ blast cells appeared in the peripheral blood. Recombinant GM-CSF was obtained for compassionate use from Schering Plough (Milan, Italy) and was administered subcutaneously twice daily at a total dose of 8 μg/kg/d, starting on day 11 from admission. Blood counts and peripheral blood smear were checked daily. There were no toxic side-effects and the disease did not progress in the peripheral blood. Due to the rapid haematological changes, GM-CSF was discontinued after 7 d. As shown in Fig 1, neutrophils rose quickly in the short-term, to fall to baseline upon GM-CSF cessation. Subsequently the granulocyte count increased steadily though slowly. A moderate decrease in the platelet count was observed during GM-CSF, followed by a rapid rise thereafter. The haemoglobin concentration never fell below 8 g/dl, the patient being transfused twice with filtered and packed red cells, and remained above 12 g/dl from the fourth week onwards. The bone marrow was checked 20 d after diagnosis, that is 2 d after completion of the GM-CSF course. Unexpectedly, the morphology of both aspirate and trephine samples was consistent with a pattern of complete remission, with no evidence of AML and restoration of normal trilineage haemopoiesis. The patient recovered totally from her pneumonia and was discharged in good health; no additional chemotherapy was given. She remained well with normal blood counts until 9 months later (July 1991) when bone marrow relapse was documented with 40% blast cells. Neutropenia was mild and there were no circulating blast cells. A second GM-CSF course was administered, using the same schedule as initially employed. Bone marrow aspirate and trephine were repeated 2 d after the end of the second GM-CSF course, demonstrating a very good partial response with 5% residual blast cells and modest dyserythropoietic changes. The patient was discharge on the 11th day from admission, and again no further therapy was planned. Although the neutrophil count increased considerably during GM-CSF administration, the subsequent haematological recovery was much slower than with the first GM-CSF course (Fig 1). Eventually the haemoglobin concentration increased to $12{\cdot}3$ g/dl, the neutrophils to $1{\cdot}7 \times 10^9$/l, and the platelet count to 238×10^9/l. In January 1992 a second overt haematological relapse was managed with a combination of mitoxantrone, cytosine

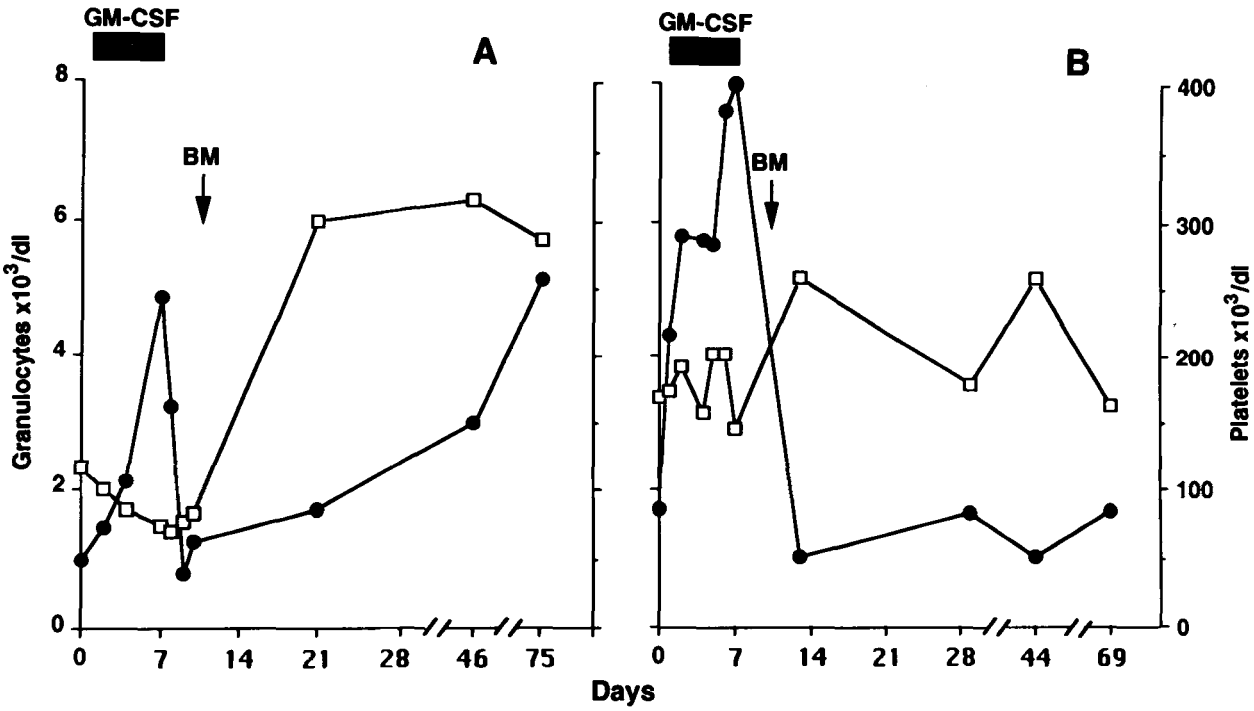

Fig 1. Early variations in granulocyte (●) and platelet (□) count during and after GM-CSF 8 μg/kg (days 1–7). BM denotes bone marrow examination. (A) GM-CSF course at presentation; (B) GM-CSF course at first relapse.

arabinoside and etoposide. A third late recurrence was treated successfully with doxorubicin and high-dose cytosine arabinoside. Currently, the patient is well and in her fourth remission, 3·5 years after acute leukaemia diagnosis, having received a myeloablative consolidation therapy with high-dose melphalan and total body irradiation supported by a peripheral blood stem cell autograft (January 1994).

DISCUSSION

We describe the very positive outcome of an adult patient with minimally differentiated or M0-AML (Bennett *et al*, 1991) and pneumonia, in whom recombinant human GM-CSF induced unexpectedly and rapidly a complete remission of considerable duration. To our knowledge this is the first response of this kind to be documented with GM-CSF. In a different therapeutic and immunobiological setting, remissions by G-CSF have been reported in patients relapsing after an allogeneic bone marrow transplantation (Giralt *et al*, 1993). What highlights our case is that a second good partial remission, albeit of shorter duration and characterized by a slower recovery from blood cytopenia, was obtained again with GM-CSF alone at first relapse 9 months later. This finding should rule out the possibility of a spontaneous remission after an infection, along with consideration that no significant change in the granulocyte count was observed in the week preceeding the first GM-CSF course. Infections usually lead to overproduction of several cytokines by monocytes, macrophages and lymphocytes (Cannistra & Griffin, 1988). Among these cytokines, TNFα and gamma-interferon were found capable of inhibiting the growth of haemopoietic cells (Broxmeyer *et al*, 1986), a mechanism perhaps explaining some spontaneous remissions in AML. However, contrary to the rapid evolutive pattern observed after GM-CSF in our case, spontaneous remissions following infections in AML were usually documented after longer time intervals since diagnosis, equal to or greater than 1 month (Paul *et al*, 1994).

Altogether, with two short GM-CSF courses, our patient spent approximately 14 months in complete remission or very good partial remission, without receiving cytotoxic therapy. The administration of GM-CSF was first considered in this infected neutropenic AML patient as an adjunct to the empirical antibiotic therapy in preparation for specific antileukaemic treatment, given that AML patients presenting with infections have a much worse prognosis (Estey *et al*, 1982). The nature of the response so promptly obtained led us to abstain from any further chemotherapy, and to observe the clinical course of the disease. The durable remissions achieved with chemotherapy at second and third relapse confirmed that prior exposure to GM-CSF did not favour the growth *in vivo* of chemoresistant cell clones.

Because leukaemic cells were not available for *in vitro* studies, we can only speculate on the possible mechanisms of response to GM-CSF. First, owing to its rapidity, we should suppose a prevalence of maturative over proliferative effects by GM-CSF on GM-CSF-sensitive AML cells, in concert with a concurrent stimulation of residual erythropoiesis and thrombocytopoiesis (Sieff *et al*, 1985). Second, we might suppose a simultaneous inhibition of clonogenic AML cell growth. The exceptional fact is that GM-CSF is a potent promoter of AML cell proliferation, resulting in disease acceleration rather than control when used prior to chemotherapy *in vivo* (Estey *et al*, 1992). However, observations from other *in vitro* and *in vivo* studies indicate that response of clonogenic AML cells to GM-CSF may not be so homogenous. For instance, GM-CSF-stimulated AML cells from one patient were reported not to grow in agar culture, suggesting involvement in terminal division (Cannistra *et al*, 1989), and the clonogenic potential of the human monoblastic U937 cell line was shown to be dramatically inhibited by GM-CSF incubation, interestingly through activation of the TNFα gene (Cannistra *et al*, 1987). Moreover, a maturation pattern with suppression of clonogenicity by GM-CSF or G-CSF was reported with the HL-60 human leukaemic cell line (Begley *et al*, 1987). In another clinical study, granulocytes increased in 2/6 AML patients treated with GM-CSF and a variable effect on AML cell cycle was observed (Preisler *et al*, 1993). In the strictly AML-related disorder myelodysplasia with blast excess, the administration of GM-CSF was sometimes associated with an increased percentage of marrow blast cells, but granulocytic responses without disease progression were more typically described (Ganser *et al*, 1989; Estey *et al*, 1991). It is worth noting that in our patient thrombocytopenia worsened during GM-CSF therapy, as observed in myelodysplastic states (Estey *et al*, 1991). The significance of this finding is unknown, but it could represent a useful hallmark of GM-CSF activity in clinical studies.

Whether GM-CSF could be considered a therapeutic agent in highly selected AML cases could be a matter of investigation. However, it must be stated that the indiscriminate use of G/GM-CSF outside a controlled research programme would be wasteful. Taken together, the current report and the data reviewed suggest that AML patients who are at increased risk of death from infection and present with few or no circulating blast cells could be considered for GM-CSF therapy, particularly when conventional treatments have failed or are contraindicated, and subject to prior *in vitro* demonstration that GM-CSF does not increase AML cell proliferation.

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