short report
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COMBINED TREATMENT WITH AMPHOTERICIN-B AND GRANULOCYTE TRANSFUSION FROM G-CSF-STIMULATED DONORS IN AN APLASTIC PATIENT WITH INVASIVE ASPERGILLOSIS UNDERGOING BONE MARROW TRANSPLANTATION

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ABSTRACT

Granulocyte transfusions from G-CSF stimulated donors were added to standard anti-infective treatment in preparation for and during allogeneic bone marrow transplantation in a young man affected by very severe acute aplastic anemia and invasive aspergillosis. Nine concentrates with a mean neutrophil content of 18.7×10^9/L (2.6×10^8/kg patient b.w.) were transfused before and after marrow infusion. An impressive clinical improvement was noticed after each granulocyte transfusion, although this was not always paralleled by a neutrophil increase in the peripheral blood. Engraftment (N >0.5×10^9/L and Plt >25×10^9/L) was verified at +16 and +40 days, respectively. The patient is currently in complete hematological and microbiological remission 14 months after transplantation. Granulocyte apheresis from G-CSF stimulated donors provides a high number of activated neutrophils. At the dose given (300 µg/day) donor tolerance to G-CSF was excellent. This new approach is indicated when life-threatening infections develop in patients exposed to prolonged severe neutropenia.

Key words: aspergillosis, granulocyte transfusion, aplastic anemia,

Acute onset of idiopathic very severe aplastic anemia (vSAA, neutrophils <0.2×10^9/L) in a young patient may require urgent transplantation.[1] Invasive aspergillosis is a life-threatening complication in aplastic patients, because antimycotic treatment is ineffective in the absence of neutrophils[2] and the presence of the infection is a serious obstacle to transplant procedures. We were able to perform a successful transplant in a young man suffering from vSAA who presented with disseminated pulmonary aspergillosis by treating the mycotic infection with amphotericin B and transfusions of granulocytes that had been harvested from G-CSF-stimulated donors.

Case Report

R. V., a 19-year-old male, was in excellent clinical condition in February 1995 when he was called up for military service.

About two weeks later, without any apparent cause, he became pale and asthenic and developed high grade fever. Admitted to a local hospital, he was found to be suffering from severe pancytopenia and bilateral pneumonia and he was transferred to our unit on February 24, 1995. A diagnosis of idiopathic vSAA (neutrophils < 0.2×10^9/L, platelets 3×10^9/L, desert bone marrow) was made, with documentation by CT scan and microbiology of rhinosinusitis and bilateral pneumonia due to *Aspergillus fumigatus* (Figure 1a).

Since the patient had an HLA-identical brother, an urgent bone marrow transplant was planned. In the meantime we treated his life-threatening invasive mycosis by combining amphotericin B (Fungizone, Squibb, 1.5 mg/kg/d from March 7, total dose 3.5 g) with granulocyte transfusions given every other day before and after transplant. Conditioning (cyclophosphamide 50 mg/kg/d for 4 days) was started on day 20 from the onset of symptoms (day 14 from diagnosis). Bone marrow cells given: 3.63×10^8/kg. GvHD prophylaxis consisted of cyclosporin A 3 mg/kg/d from days –1 to +20 iv, then 10 mg/kg/d orally for one year. The patient also received GM-CSF (Molgramostim, Schering-Plough, 300 mg/d from days +1 to +30) and G-CSF (Filgrastim, Amgen, 300 mg/d from days +9 to +30), itraconazole (400 mg/d from day +27), acyclovir (from days –7 to +30), polyspecific (30 g twice weekly) and anti-CMV (100 mL, twice weekly) IgG. Time to neutrophil (> 0.5×10^9/L) and

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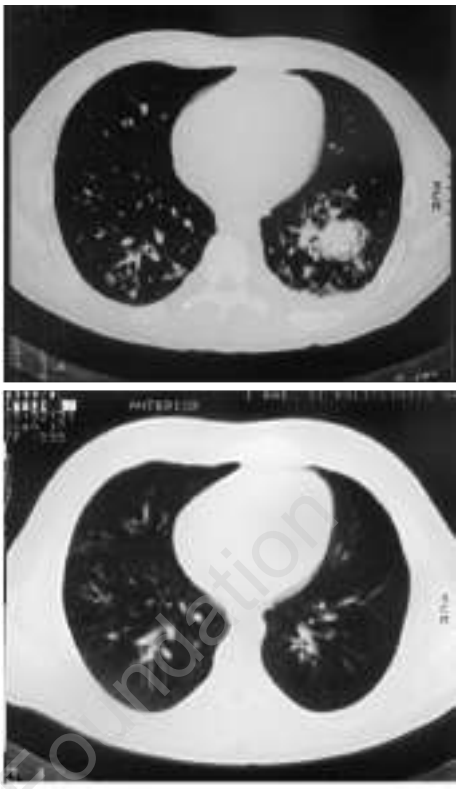
platelet (>25×10⁹/L) recovery was +16 and +40 days, respectively. Microbiology became negative within two weeks. At day +46 a repeat amphotericin B course (100 mg every other day, total 3 g) was required because of a recurrence of fever and the presence of aspergillus in the sputum. No signs of acute or chronic graft versus host disease appeared during the post-transplant period. In October 1996, 19 months after transplantation, the patient is in complete hematological and microbiological remission (Figure 1b).

Granulocyte apheresis from G-CSF-stimulated donors

Nine granulocyte concentrates obtained from three donors were infused over two weeks. Aphereses were performed with relatives of the patient (including the bone marrow donor) who shared the same blood group; the patient and the donors were all positive for anti-CMV IgG. In order to avoid presensitization against donor antigens, the bone marrow donor was utilized as a granulocyte donor only after transplantation. After giving informed consent, each donor received 300 µg of r-metHuG-CSF (Filgrastim, Amgen) once a day for 5 days. Their circulating neutrophils immediately peaked between 30 and 40×10⁹/L; moderate bone pain was the only complaint. Apheresis procedures were performed every other day and were well tolerated. Mean bag content was 18.7×10⁹ neutrophils. Each bag was irradiated before infusion.



A

B

Figure 1. A: an excavated lesion caused by *Aspergillus fumigatus* in the left lung, before transplant. B: Complete recovery five months later.

Comment

Aspergillosis is usually a late complication of AA, often triggered by immunosuppressive treatments; in our patient invasive aspergillosis was diagnosed simultaneously with vSAA before any treatment was given. An aplastic patient with invasive aspergillosis is the worst possible candidate for a bone marrow trasplant. A recent review[3] reports that only 10% of aplastic patients with invasive aspergillosis respond to standard anti-fungal treatment. Transfusion of granulocytes to prevent or treat infection in neutropenic patients was studied in the 70's.[4] Reduction of bacterial infections and mortality were described, but harvest yield was low and concern was raised about the possibility of pulmonary damage.[5] Nowadays, the availability of colony-stimulating factors has increased the neutrophil yield up to six times, without significant side-effects in the donor;[6] neutrophil function in the harvest is fully retained or even increased.[7] This has triggered new interest in granulocyte transfusion.[8]

The favorable outcome in our patient suggests that granulocyte transfusion should be included in the treatment strategy for such patients. Lung sequestration is a well-known phenomenon after granulocyte infusion,[9] which in this case may have favored the resolution of the pneumonia. Also, it is possible that neutrophil activation by growth factors played some role in the antimycotic defense.[10]

References

1. Bacigalupo A. Guidelines for the treatment of severe aplastic anemia. Haematologica 1994; 79:438-44.
2. Saral R. Candida and Aspergillus infections in immunocompromised patients. Rev Infect Dis 1991; 13:487-92.
3. Denning DW. The treatment of invasive aspergillosis. J Infect 1994; 28 (suppl 1):25-33.
4. Clift RA, Sanders JE, Thomas ED, Williams B, Buckner CD. Granulocyte transfusions for the prevention of infection in patients receiving bone marrow transplants. N Engl J Med 1978; 298:1052-7.
5. Bensinger WI, Price TH, Dale DC, et al. The effects of daily recombinant human granulocyte colony-stimulating factor administration on normal granulocyte donors undergoing leukapheresis. Blood 1993; 81:1883-8.
6. Caspar CB, Seger RA, Burger J, Gmur J. Effective stimulation of donors for granulocyte transfusions with recombinant methionyl granulocyte colony-stimulating factor. Blood 1993; 81:2866-71.
7. Katoh M, Takada M, Nakatoni N, Cho C, Umeda M. Chemotaxis and phagocytosis of the neutrophils mobilized by granulocyte colony-stimulating factors in healthy donors for granulocyte transfusion. Am J Hematol 1995; 49:96-7.
8. Freireich EJ. White cell transfusions born again. Leuk Lymphoma 1993; 11(Suppl 2):161-5.
9. Strauss RG, Connett JE, Gale RP, et al. A controlled trial of prophylactic granulocyte transfusions during initial induction chemotherapy for acute myelogenous leukemia. N Engl J Med 1981; 305:597-602.
10. Vannucchi AM, Bosi A, Laszlo D, Guidi S, Saccardi R, Rossi Ferrini P. Treatment of a delayed graft failure after allogeneic bone marrow