Complete Remission Following Chemotherapy with Low-dose Cytosine Arabinoside and Macrophage Colony-stimulating Factor/Granulocyte Colony-stimulating Factor in a Patient with Relapsed Acute Myeloid Leukemia after Stem Cell Transplantation

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Abstract

The prognosis of patients who relapse with acute myeloid leukemia (AML) after undergoing stem cell transplantation (SCT) is poor. There exist some treatments for relapsed AML; however, almost all treatments are associated with a high level of regimen-related toxicities (RRTs). The RRT of donor lymphocyte infusion is lower than that of other treatments; however, the efficacy of this treatment in treating patients with relapsed AML is lower than that observed in patients with chronic myelomonocytic leukemia. We herein report a case of relapsed AML after SCT in a 65-year-old man. We performed donor lymphocyte infusion; however, it was not effective. We then administered chemotherapy with cytosine arabinoside and macrophage colony-stimulating factor/granulocyte colony-stimulating factor and complete remission was achieved. Since graft-versus-host disease occurred after the administration of low-dose chemotherapy in this case, we speculated that the chemotherapy induced a graft-versus-leukemia effect.

Key words: acute myeloid leukemia, allogeneic stem cell transplantation, chemotherapy, cytokines, donor and therapeutic apheresis, graft-versus-tumor effect

Case Report

A 65-year-old man was diagnosed as having acute myeloid leukemia (AML) (FAB: M6) in 2005. Complete remission (CR) was achieved using chemotherapy with idarubicin and cytosine arabinoside (AraC). The patient underwent two courses of consolidation with mitoxantrone and AraC and enocitabine, etoposide and vincristine. Bone marrow transplantation (BMT) from an HLA one-locus mismatched unrelated donor (HLA-B) was performed after the first CR was achieved. Fludarabine (30 mg/m² on days -7-2), busulfan (4 mg/kg on days -3 and -2) and total body irradiation at 4 Gy (day -1) were administered for conditioning. The total infused dose of mononuclear cells was 3.25×10⁸/kg. Cyclosporin and short-term methotrexate were administered as prophylaxis for acute graft-versus-host disease (GVHD). Engraftment with an absolute neutrophil count of more than 500/μL was seen on day 16. Bone marrow aspiration obtained on day 22 showed complete molecular remission, as the expression of WT-1 mRNA was within the normal range. Acute GVHD, grade II (skin stage 3) was observed; however, no chronic GVHD occurred. The patient was discharged on day 87. His bone marrow was periodically checked and he was confirmed to be in CR.

Bone marrow aspiration performed 18 months after BMT due to a decrease in the platelet count showed an early relapse of AML (4.2% blasts). Bone marrow aspiration performed weekly for three weeks after relapse showed non-progressive findings. No clinical GVHD was observed at

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that time. The patient was hospitalized to undergo donor lymphocyte infusion (DLI) on day 564 (Table).

CD3-positive lymphocytes obtained with leukapheresis from the same donor were infused. The first DLI was performed on day 591 after BMT at a dose of 1.0×10<sup>7</sup>/kg, the second was performed on day 13 after the first DLI at a dose of 2.0×10<sup>7</sup>/kg and the third was performed on day 27 at a dose of 1.87×10<sup>7</sup>/kg. Bone marrow aspiration performed after the first and second DLIs showed no suppressive effects on AML cells (5.7% and 8.1%, respectively). However, bone marrow aspiration performed after the third DLI showed an increase in the percentage of AML cells (18.6%) (Fig. 1). No clinical GVHD was observed at that time. Therefore, on day 630 after BMT, the patient was treated
with low-dose AraC and macrophage colony-stimulating factor (M-CSF) (AraC at a dose of 24 mg/body ×2 on days 1-3 and then reduced to 20 mg/body ×2 on days 3-14 due to febrile neutropenia (FN) and M-CSF at a dose of 800×10⁴ units on days 14-21) (Fig. 2). Later, *Escherichia coli* (*E. coli*) was detected in a blood culture. The sepsis improved; however, the neutropenia persisted. On day 21 after starting chemotherapy, FN again developed. We therefore switched from M-CSF to granulocyte colony-stimulating factor (G-CSF). On day 35 after starting chemotherapy, the hematopoiesis recovered and bone marrow aspiration showed a decrease in the percentage of AML cells (1.5%). However, a skin eruption, which had persisted since day 7 after starting chemotherapy (day 46 after the first DLI), spread to the whole body on day 43 after starting chemotherapy (day 82 after the first DLI). At that time, the levels of liver enzymes and bilirubin were elevated. Skin and liver biopsies were performed, and the patient was diagnosed as having GVHD. The GVHD was compatible with acute type grade III disease (skin stage 3, liver stage 2, gut stage 0). The interleukin (IL)-1beta, IL-6, interferon (IFN)-gamma and tumor necrosis factor (TNF)-alpha levels were elevated after chemotherapy and before the onset of GVHD (Fig. 3). The hematopoiesis recovered and the AML cells disappeared in the bone marrow after GVHD was diagnosed. Prednisolone and cyclosporine were administered to treat the GVHD. The skin eruption disappeared and the liver enzyme levels decreased. However, the high bilirubin level (T-bil: 2 mg/dL) persisted. The patient was discharged on day 716 after BMT (day 125 after the first DLI) (Fig. 2). The bilirubin level remained above the normal range (1-2 mg/dL) for approximately 20 months after the first DLI. Molecular CR, the patient’s minimal residual disease being evaluated using the WT-1 mRNA level, persisted for 30 months after the first DLI.

**Discussion**

The prognosis of patients who relapse with AML after undergoing stem cell transplantation (SCT) is poor (1). There exist some treatments for relapsed AML such as discontinuing immunosuppressants, donor lymphocyte infusion, chemotherapy and re-transplantation (1). However, the regimen-related toxicity and treatment-related mortality of high-dose chemotherapy and re-transplantation are often very high (1). We therefore decided to perform DLI because the burden of AML was small and growth was slow at that time. Although we performed DLI, it was not effective for the patient. The AML rapidly worsened after the last DLI was performed. Next, we administered low-dose AraC and M-CSF therapy because this therapy is safer than high-dose chemotherapy. Surprisingly, GVHD occurred at almost the same time as recovery of the patient’s bone marrow function. Then, the tumor burden decreased. Finally, CR was achieved. Considering the patient’s clinical course, we believe that CR was achieved in this case due to an additional graft-versus-leukemia (GVL) effect rather than the cytoreduction achieved with chemotherapy alone.

We speculate that three factors induced the GVL effect that occurred after therapy. The first factor involves tumor-associated proteins. It is known that a large amount of tumor-associated proteins, such as tumor antigens and ligands of toll-like receptors (TLRs), is released by massive
and radiation. Additionally, tumor-associated proteins have been found to activate tumor-associated antigen-presenting cells and effector T cells in a mouse model (2, 3).

The second factor is the administration of M-CSF and G-CSF. M-CSF generates myeloid cells, such as monocytes and macrophages, both of which induce the production of many inflammatory cytokines. It has been reported that CR is achieved with G-CSF monotherapy in patients with relapsed AML or myelodysplastic syndrome occurring after transplantation (4). The last factor is E. coli infection during myelosuppression. E. coli lipopolysaccharides bind to TLR4 and induce innate immunity. They also play an important role in the onset of acute GVHD induced by conditioning therapy for bone marrow transplantation (5).

We measured the concentrations of inflammatory cytokines in order to determine whether inflammatory cytokines affected the patient’s clinical course. As shown in Fig. 3, the levels of IL-1beta, IL-6, IFN-gamma and TNF-alpha were elevated and IL-10 was not detected, as previously reported (6, 7). Although we could not determine the detailed mechanisms by which the hematopoietic cells were completely replaced with donor-derived cells from the mixed chimera after chemotherapy, it is possible that some or all of the therapies and events in this case induced GVL.

DLI is an immunotherapy used to strengthen the GVL effect. It is known that DLI is effective for treating relapsed chronic myelomonocytic leukemia after SCT, but it is less effective for treating AML due to the high proliferation rate of leukemic cells and the large tumor burden at the time of relapse. Although chemotherapy is added to DLI for the purpose of achieving cytoreduction in some studies, chemotherapy may also induce tumor-associated proteins. The clinical course of our patient suggests that low-dose chemotherapy may induce not only cytoreduction, but also GVL effects. The use of cytokine therapies including DLI to strengthen tumor immunity has been reported (8-10). Low-dose chemotherapy with DLI may be as effective as these cytokine therapies for treating relapsed AML after SCT.

The authors state that they have no Conflict of Interest (COI).

References

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