Successful Allogeneic Hematopoietic Stem Cell Transplantation for Aggressive NK Cell Leukemia

Satoshi Ichikawa, Noriko Fukuhara, Joji Yamamoto, Makiko Suzuki, Shinji Nakajima, Yoko Okitsu, Katsura Kohata, Yasushi Onishi, Kenichi Ishizawa, Junichi Kameoka and Hideo Harigae

Abstract

Aggressive natural killer cell leukemia (ANKL) is a highly aggressive lymphoproliferative disease. An appropriate therapeutic strategy for ANKL remains to be established, but a few case reports have suggested that allogeneic hematopoietic stem cell transplantation (allo-HCT) can be curative. Here, we report a young woman with ANKL showing central nervous system (CNS) invasion, who has been in complete remission for more than a year after allo-HCT following two courses of intravenous chemotherapy and several rounds of intrathecal chemotherapy. Intensive remission induction chemotherapy followed by conventional myeloablative allo-HCT is a promising approach for long-term remission in cases of this aggressive malignancy.

Key words: Aggressive NK cell leukemia, allogeneic hematopoietic stem cell transplantation, central nervous system invasion, subarachnoid hemorrhage, L-asparaginase


Introduction

Aggressive natural killer cell leukemia (ANKL) is a malignant lymphoproliferative disease defined as a systemic neoplastic proliferation of natural killer (NK) cells associated with Epstein-Barr virus (EBV) and a fulminating clinical course (1). ANKL is a rare form of leukemia, which is most prevalent among Asian populations.

ANKL has several clinicopathological characteristics (1-3). Leukemic cells are morphologically slightly immature lymphocytes with large granules, and are immunophenotypically CD2+ surface-CD3- cytoplasmic-CD3ε+/ CD16+/- CD56+ CD57-, without rearrangement of the T-cell receptor or immunoglobulin heavy chain gene. Monoclonality of EBV is usually detected in the peripheral blood and/or bone marrow. The disease is systemic, and marked liver dysfunction and massive hepatosplenomegaly are often detected.

The prognosis of ANKL is extremely poor. The median survival from diagnosis of ANKL is reported to be less than 2 months, which is equivalent to that of advanced extranodal NK-cell lymphoma, nasal type (ENKL) (2, 4). ANKL may be a leukemic counterpart of ENKL, although there are some clinicopathological differences between the two diseases (1, 2). Standard therapy for ANKL has not been established, although some case reports have shown successful outcomes of allogeneic hematopoietic stem cell transplantation (allo-HCT) (5, 6).

Here, we report a case of ANKL in a young woman. We performed treatment with aggressive induction chemotherapy and allogeneic peripheral blood stem cell transplantation (allo-PBSCT) from an HLA-matched sibling within 2 months after diagnosis. The patient has been in remission for one year since transplantation. This approach represents a promising therapeutic strategy for ANKL; strong remission induction by aggressive chemotherapy followed sequentially by allo-HCT.

Case Report

A 28-year-old woman with fever and abdominal disten-
A patient with leukemic large granular lymphocytes was diagnosed with ALK+ anaplastic large cell lymphoma (ANKL) after presenting with hepatosplenomegaly and liver dysfunction. The patient was admitted to Tohoku University Hospital for further evaluation and treatment.

**Laboratory Data at Presentation**

<table>
<thead>
<tr>
<th>Complete blood count</th>
<th>Biochemistry</th>
<th>EBV serological status</th>
</tr>
</thead>
<tbody>
<tr>
<td>WBC 8100 /μL Neut 10 % Mono 4 % *Lym 85 %</td>
<td>T-BIL 7.3 mg/dL D-BIL 5.4 mg/dL ALP 2073 IU/L γ-GTP 82 IU/L</td>
<td>anti-VCA IgG ×1280 anti-VCA IgM ×10 anti-EBNA Ab ×320</td>
</tr>
<tr>
<td>RBC 3.74 ×10¹²/μL Hb 9.5 g/dL HCT 29 %</td>
<td>AST 254 IU/L ALT 251 IU/L LDH 1190 IU/L</td>
<td>EBV-DNA (PB) 2.3 ×10⁵ copies/μgDNA</td>
</tr>
<tr>
<td>PLT 10.0 ×10⁹/μL Ret 2.1 %</td>
<td>BUN 8 mg/dL Cr 0.4 mg/dl UA 3.4 mg/dl sIL-2R 2977 IU/L</td>
<td>EBV-DNA southern blot Monoclonality+</td>
</tr>
<tr>
<td>Coagulation</td>
<td>Haptoglobin &lt; 6.9 mg/dL Ferritin 1197 ng/mL CRP 0.1 mg/dL</td>
<td></td>
</tr>
<tr>
<td>PT-INR 1.13 APTT 60.9 sec</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fibrinogen 74 mg/dL FDP 2.5 μg/mL D-dimer 1.0 μg/mL</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*89% of lymphocytes showed NK cell phenotype defined by flow cytometry.*

Bone marrow showed monotonous proliferation of large lymphocytes, which had cytoplasmic azurophilic granules (Fig. 1). Marked liver dysfunction and coagulatory disturbance can be due to massive infiltration of leukemic cells into the liver. Flow cytometry revealed a population of abnormal lymphocytes that were CD2⁻ surface-CD3⁻CD16⁺CD56⁺CD57⁻. Viral load of Epstein-Barr virus (EBV-VL) in whole blood, measured by quantitative real-time polymerase chain reaction (RT-PCR) of EBV DNA, was extremely high (Table 1), and monoclonality of infected EBV was confirmed by Southern blotting analysis (data not shown). RT-PCR of EBV DNA was performed as previously reported (7) with minor modifications. Spinal tap was not initially performed due to coagulatory disturbance. A diagnosis of ANKL was made based on the above data, and chemotherapy was initiated with CHOP regimen (cyclophosphamide, doxorubicin, vincristine, and prednisone) to reduce tumor burden. The administration of danaparoid was started to control coagulatory disturbance. On the third day of the CHOP regimen, the patient suddenly suffered from incomplete paralysis of the left oculomotor nerve. Brain CT revealed subarachnoid hemorrhage (SAH) at the left side of the suprasellar cistern (Fig. 2). Cerebral angiography could not detect causative lesions, such as arteriovenous malformation or aneurysm. The administration of danaparoid was stopped immediately, and SAH showed no further exacerbation. Two weeks later, coagulatory function recovered to within the normal range, and spinal tap was performed. Cytology of cerebrospinal fluid (CSF) detected abnormal lymphocytes, and EBV-VL of CSF was high (2.3×10⁵ copies/μg DNA), suggesting invasion of leukemic cells. The second chemotherapy regimen consisting of L-asparaginase (L-asp), methotrexate (MTX), etoposide, ifosfamide, and steroids, was initiated 2 weeks after the CHOP regimen. Intrathecal chemotherapy consisting of a combination of MTX, cytarabine, and pred-

---

**Table 1. Laboratory Data at Presentation**

<table>
<thead>
<tr>
<th>Complete blood count</th>
<th>Biochemistry</th>
<th>EBV serological status</th>
</tr>
</thead>
<tbody>
<tr>
<td>WBC 8100 /μL Neut 10 % Mono 4 % *Lym 85 %</td>
<td>T-BIL 7.3 mg/dL D-BIL 5.4 mg/dL ALP 2073 IU/L γ-GTP 82 IU/L</td>
<td>anti-VCA IgG ×1280 anti-VCA IgM ×10 anti-EBNA Ab ×320</td>
</tr>
<tr>
<td>RBC 3.74 ×10¹²/μL Hb 9.5 g/dL HCT 29 %</td>
<td>AST 254 IU/L ALT 251 IU/L LDH 1190 IU/L</td>
<td>EBV-DNA (PB) 2.3 ×10⁵ copies/μgDNA</td>
</tr>
<tr>
<td>PLT 10.0 ×10⁹/μL Ret 2.1 %</td>
<td>BUN 8 mg/dL Cr 0.4 mg/dl UA 3.4 mg/dl sIL-2R 2977 IU/L</td>
<td>EBV-DNA southern blot Monoclonality+</td>
</tr>
<tr>
<td>Coagulation</td>
<td>Haptoglobin &lt; 6.9 mg/dL Ferritin 1197 ng/mL CRP 0.1 mg/dL</td>
<td></td>
</tr>
<tr>
<td>PT-INR 1.13 APTT 60.9 sec</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fibrinogen 74 mg/dL FDP 2.5 μg/mL D-dimer 1.0 μg/mL</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*89% of lymphocytes showed NK cell phenotype defined by flow cytometry.*
nisolone, was also performed five times on a weekly basis. After l-asparaginase-containing second chemotherapy, liver dysfunction was remarkably improved and EBV-VL of peripheral blood (PB) and CSF fell to below the level of detection (Fig. 3).

Considering the extreme aggressiveness of ANKL, we performed allogeneic peripheral blood stem cell transplantation (allo-PBSCT) from an HLA-matched sibling about 2 months after the initiation of treatment. After a conditioning regimen consisting of total-body irradiation (2 Gy ×6) and high-dose cyclophosphamide (60 mg/kg/day, for 2 days), 4.9×10^6/kg of CD34+ cells were infused. Cyclosporin A (CsA) and short-term MTX were used as prophylaxis for graft-versus-host disease (GVHD). Neutrophil engraftment was confirmed on day 12 without severe regimen-related toxicity. There were no apparent signs of engraftment syndrome or acute GVHD. Oculomotor paralysis caused by SAH disappeared completely after allo-PBSCT. CsA was tapered gradually from around day 30, and stopped on day 105. Mild chronic GVHD of oral mucosa emerged shortly after the discontinuation of CsA, but was controllable without general administration of steroids or any additional immunosuppressants. EBV-VL of PB and CSF has been below the level of detection for more than one year after allo-PBSCT (Fig. 3), and ANKL is considered to be in complete remission (CR) in this patient.

**Discussion**

We successfully treated a young female ANKL patient complicated by CNS invasion with an intensive chemotherapeutic regimen and conventional myeloablative allo-HCT. ANKL is usually refractory to chemotherapy, which can be partially explained by the expression of P-glycoprotein.

Figure 2. Cranial computed tomography scan at the onset of subarachnoid hemorrhage. The arrow indicated the hemorrhage at the left side of the suprasellar cistern.

Figure 3. Epstein-Barr virus (EBV) viral load. After chemotherapy with l-asparaginase-containing regimen, EBV viral load in whole blood and cerebrospinal fluid fell to below the level of detection, which has been maintained until one year after transplantation. CHOP, cyclophosphamide, doxorubicin, vincristine, and prednisone; IT, intrathecal chemotherapy with methotrexate, cytosine arabinoside, and prednisolone; l-asp, l-asparaginase; PB, peripheral blood; BM, bone marrow, CSF, cerebrospinal fluid; Sib/PBSCT, allogeneic peripheral blood stem cell transplantation from an HLA-matched sibling donor; cGVHD, chronic graft-versus-host disease.
(P-gp) (3, 9), a multidrug resistance gene-encoded protein. L-Asp is thought not to be a substrate of P-gp, and combination chemotherapy including L-asp was reported to be effective for ANKL and ENKL (8, 10, 11). Consistent with these reports, an L-asp-containing chemotherapeutic regimen was highly effective in the present case, leading to good disease status before allo-HCT.

ANKL is hardly curable by chemotherapy and the average duration of survival after diagnosis is less than one year, but long-term survival after allo-HCT has been reported. Ito et al reported three cases of ANKL (5); two were treated with cord blood stem cell transplantation, and the other was treated with bone marrow transplantation from a sibling donor. All cases were in partial response (PR) before allo-HCT, and achieved CR after allo-HCT. Suzuki et al reported two cases of ANKL (6); one patient was primarily refractory to chemotherapy and died despite myeloablative allo-HCT, and the other achieved CR after allo-PBSCT. Due to the rarity of ANKL, a standard therapeutic strategy remains to be determined. However, these reports and the clinical course of the present case suggest that conventional myeloablative allo-HCT in patients with good disease status may lead to long-term survival.

In the present case, high EBV-VL was detected in PB and CSF at presentation. However, it became undetectable after the second course of chemotherapy, and has not been detected for more than one year since allo-HCT, consistent with the good clinical course. This result is consistent with previous reports, which suggest that quantification of EBV-VL in EBV-related NK-cell neoplasms could be a useful surrogate marker of tumor load (5, 12, 13). In addition, this report may be informative from the point that quantification of EBV-VL is useful to evaluate CNS involvement, which was hardly evaluated in previous reports.

The only major complication occurring during the treatment was SAH. Intracranial hemorrhage (ICH) is sometimes seen in patients with hematological malignancy (14, 15). However, SAH in the present case was relatively mild and was resolved without neurological sequelae. ICH in hematological malignancy may be correlated with several risk factors, such as vessel wall anomalies, low platelet counts, platelet dysfunction, coagulation factor deficiency, disseminated intravascular coagulation, and hyperleukocytosis (14, 16). In the present case, vessel wall anomalies were not detected by radiographic studies. Coagulatory disturbance and low platelet counts could be causative of SAH. However, the possibility of microvessel rupture by meningeal infiltration of leukemic cells could not be excluded, because symptoms were mild and improved spontaneously after chemotherapy.

In summary, intensive remission induction by aggressive chemotherapy followed by early myeloablative allo-HCT represents a promising therapeutic strategy for ANKL. Clinical trials should be performed to establish the optimal therapeutic strategy for ANKL.

References