Autologous peripheral blood stem cell transplantation in children and adolescents with non-Hodgkin lymphoma

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Abstract. The aim of this study was to evaluate the effect and safety of autologous peripheral blood stem cell transplantation (APBSCT) in children and adolescents with non-Hodgkin lymphoma (NHL). Ten patients with NHL were analyzed retrospectively. In all the patients, lymph node enlargement was most frequently detected. Patients with a mediastinal mass presented with a cough, palpitation and shortness of breath. Extranodal patients presented with abdominal pain, inability to walk and vaginal bleeding. All patients underwent APBSCT with conditioning regimens BEAM or BuCy. Among them, four patients with B-cell NHL received rituximab in addition to the conditioning regimen. Hematopoietic reconstitution was observed in all patients. Severe toxicity and transplant-related mortality were not observed. Prior to APBSCT, nine patients with a status of complete response (CR) and CR unconfirmed achieved continuing complete remission. Only one patient with partial response succumbed to progressive disease. APBSCT in children and adolescents with NHL is a safe, convenient and efficient treatment. The BEAM conditioning regimen was shown to be effective and tolerable for children and adolescents with NHL. Rituximab is a safe agent in the transplantation. The CR status at the time of transplantation demonstrated a higher survival rate.

Introduction

Non-Hodgkin lymphoma (NHL) is the third most common cancer (10%), accounting for ~60% of all lymphomas in children and adolescents (1). The most common symptoms of pediatric NHL are fever, superficial lymph node enlargement, cough, shortness of breath, anorexia, abdominal pain, sore throat and nasal obstruction. The major histological subtypes of NHL include lymphoblastic lymphoma (LBL),oBurkitt lymphoma (BL), anaplastic large-cell lymphoma and diffuse large B-cell lymphoma (2). Recent developments in chemotherapy regimens, including BFM90, VDCLP and HyperCVAD/MA, intrathecal methotraxate, cytarabine, dexamethasone and combined chemotherapy with rituximab for B-cell NHL patients, have rapidly improved the survival rate of pediatric NHL patients (3-6). Long term event-free survival (EFS) has been exhibited to be 60-90%. However, for ~10-30% of children with NHL who receive modern chemotherapy, the treatment is likely to be unsuccessful (7-10). Recently, autologous peripheral blood stem cell transplantation (APBSCT), with conditioning regimens BEAM or BuCy, has improved the long-term survival of children with NHL (4,9). A previous study revealed that the 3-year EFS in pediatric NHL patients receiving APBSCT was 70% (7). In another study, pediatric patients with refractory or relapsed NHL underwent APBSCT with conditioning regimens of busulfan and cyclophosphamide and exhibited a disease-free survival rate of 90% (11). In the present study, we explored the use of APBSCT in children and adolescents with NHL. In total, 9 patients with a complete response (CR) status prior to APBSCT survived without disease. Transplantation-associated mortality was not observed. Furthermore, the present study discussed 4 representative NHL cases in more detail.

Patients and methods

Patients. The study was approved by the ethics committee of Shanxi Tumor Hospital (Taiyuan, China). Between January 2008 and December 2013, 10 children and adolescents with NHL underwent APBSCT at the Department of Hematology, Shanxi Tumor Hospital. NHL was diagnosed according to the 2008 World Health Organization criteria (12) and all biopsy specimens were re-evaluated in our institution. The patients were followed-up from diagnosis until mortality or the last follow-up on March 31st, 2015. The patients comprised eight males and two females. The median age was 16 years (range, 10-20 years). Most of the patients had aggressive lymphoma and were in stage III/IV. Six patients were in first complete remission (CR_1), two were in CR_2, one had unconfirmed complete remission (CR_u) and one had a partial response (PR) prior to APBSCT (Table I). Written informed consent was obtained from the patients' families prior to participation in the study.

Treatment. Peripheral blood progenitor cells were mobilized by chemotherapy and granulocyte colony stimulating factor

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Table I. Characteristics of patients with non-Hodgkin lymphoma at transplantation.

Patient no.	Age/ gender	Sites of disease	Stage	aaIPI	BM (blast cells, %)	Pathological type	Status before APBSCT	Conditioning regimen	Outcome	OS (months)
1	11/F	Thoracic vertebral	IVA	2	-	B-LBL	CR_1	R-BEAM	Alive with NED	45
2	11/M	Ileocecum	IIIA	2	-	MALToma	CR2	BEAM	Alive with NED	78
3	10/M	Inguinal lymph node	IVA	2	84.5	B-LBL	CR1	R-BuCy	Alive with NED	46
4	15/M	Cervical lymph mode, mediastinum, pleural and pericardial effusion and splenomegaly	IVA	2	4.5	T-LBL	CR1	BEAM	Alive with NED	26
5	20/M	Cervical lymph nodes	IIA	0	-	B-LBL	CR_1	R-BEAM	Alive with NED	44
6	19/M	Stomach, abdominal lymph nodes and splenomegaly	IIIA	2	-	GZL	CR_1	BEAM	Alive with NED	29
7	20/M	Mediastinum, pleural and pericardial effusion, ascites, cervical and abdominal lymph nodes	IVB	3	-	DLBCL	CR_u	BEAM	Alive with NED	17
8	14/M	Mediastinum, pleural effusion and cervical lymph nodes	IVB	3	4.0	T-LBL	PR	BEAM	Dead with PD	6
9	17/M	Pharynx, tonsil, cervical lymph nodes and splenomegaly	IIIB	1	1.5	NKTCL	CR_1	BEAM	Alive with NED	34
10	20/F	Ovary, liver, spleen, BM and CNS	IVB	3	75.0	DLBCL	CR_2	R-BEAM	Alive with NED	29

aaIPI, age-adjusted international prognostic index; BM, bone marrow; OS, overall survival; APBSCT, autologous peripheral blood stem cell transplantation; CNS, central nervous system; B-LBL, B-cell lymphoblastic lymphoma; GZL, gray zone lymphoma; DLBCL, diffuse large B-cell lymphoma; T-LBL, T-cell lymphoblastic lymphoma; NKTCL, natural killer T-cell lymphoma; MALToma, mucosa-associated lymphoid tissue lymphoma; CR_1, first complete remission; CR_2, second complete remission; CR_u, unconfirmed complete remission; PR, partial remission; R-BEAM, BEAM plus rituximab; NED, no evidence of disease; PD, progressive disease.

(G-CSF; Kyowa Hakko Kirin Co., Ltd., Tokyo, Japan). Peripheral blood stem cells (PBSCs) were harvested using a standard procedure on a COBE spectra apheresis system (Terumo BCT, Inc., Lakewood, CO, USA) 2-4 times. The cells were cryopreserved in -80˚C refrigerate (Sanyo Electric Co., Ltd., Moriguchi, Japan). Cells were successfully collected from all patients. The median mononuclear cell dose infused was $7.8x10^8$/kg (range, 6.61-12.38x10^8/kg) and the median $CD34^+$ cell dose infused was $4.81x10^6$/kg (range, 1.09-12.13x10^6/kg). Hematopoietic reconstitution was observed in all patients. The median time to neutrophil engraftment was 10 days (range, 8-14 days) and to platelet engraftment was 15 days (range, 12-19 days). Following APBSCT, two patients with bulky mediastinal mass received local radiotherapy and one patient with central nervous system involvement received craniospinal radiotherapy. Nine patients with CR_1, CR_2 and CR_u survived with no evidence of disease. Only one patient with PR succumbed to progressive disease.

Conditioning regimen. Nine patients received the BEAM regimen (carmustin 300 mg/m^2x1, day -6; etoposide 75 mg/m^2, Q12hx4, days -5, -4, -3 and -2; cytarabine 100 mg/m^2, Q12hx4, days -5, -4, -3 and -2; and melphalan 140 mg/m^2x1, day -1). One patient received the modified BuCy regimen (hydroxyurea 2 g/m^2, Q12hx2, days -9 and -8; cytarabine 2 g/m^2x1, day -7; busulfan 1 mg/kg, Q6hx2, days -6 and -5; cyclophosphamide 1.8 g/m^2x2, days -4 and -3; and lomustine 0.2 g/m^2x1, day -2). Four patients received rituximab 375 mg/m^2x2, days -1 and +8.

Case 1. An 11-year-old female sought care at the Second Affiliated Hospital of Shanxi Medical University in July 2011 with a 2-month history of back pain, progressive weakness of the legs and the inability to walk. Magnetic resonance imaging demonstrated a thoracic vertebral (T_6) mass with spinal cord compression. A bone scan revealed no other bone abnormalities. The ultrasound presented indications of hypo-echoic hepatic lesions, mild splenomegaly, and no superficial lymph node enlargement. Results of a bone marrow (BM) smear and bone marrow biopsy (BMB) were normal. Flow cytometry (FCM) of BM cells revealed that 12.5% of cells were abnormal. Cells were positive for CD10, CD19, $CD79_a$ and CD20, and negative for CD34. The patient underwent surgical resection of the T_6 vertebral mass. Titanium mesh implantation and a bone graft were used for internal fixation. Histological examination revealed small to medium mononuclear cells with conspicuous nucleoli. Immunohistochemical (IHC) staining revealed positivity for LCA, CD99, TdT, $CD79_a$, PAX5 and CD20, Ki67 90%$^+$, and negativity for CD3, CD34 and MPO. The diagnosis of B-cell lymphoblastic lymphoma (B-LBL) was assigned. The patient received the BFM 90 protocol and achieved CR. Two months after the end of treatment, the patient was admitted to our hospital in June 2012 for APBSCT. Physical examination revealed the ability to walk, no superficial lymph node enlargement and hepatosplenomegaly. Laboratory tests revealed a hemoglobin (HGB) level of 98 g/l, white blood cell (WBC) count of 1.7x10^9/l, platelet (PLT) count of 36x10^9/l, and lactate dehydrogenase (LDH) of 171 U/l. The liver and renal functions, total protein levels, a coagulation test, and cerebrospinal fluid (CSF), cytomegalovirus (CMV) and Epstein-Barr virus (EBV) tests were normal. A BM smear and BMB were normal. Ultrasound results were unremarkable. A positron emission tomography-computed tomography (PET-CT) scan revealed no evidence of a tumor. The child was finally diagnosed as having primary thoracic vertebral B-LBL stage IVA with an age-adjusted international prognostic index (aaIPI) of 2 and an Eastern Cooperative Oncology Group (ECOG) score of 2. Four months later, the patient underwent APBSCT. The conditioning regimen was BEAM plus rituximab (R-BEAM). Twenty-nine months after APBSCT, she is now well and in continuing complete remission (CCR).

Case 2. An 11-year-old male sought care in an outside hospital with a 1-week history of abdominal pain. Ultrasound revealed a mass in the right lower quadrant. Exploratory laparotomy was performed. A 5x5-cm mass was resected, and an ileocecostomy was carried out. Histopathological examination revealed the aggregation of small atypical lymphoid cells. IHC staining revealed positivity for LCA, CD20 and $CD79_a$, Ki67 40%$^+$, and negativity for CD3, CD5, CD23 and cyclin D1. The diagnosis was mucosa-associated lymphoid tissue lymphoma (MALToma).

The patient was admitted to our hospital in September 2008 for chemotherapy. Physical examination revealed a good state, no superficial lymph node enlargement or hepatosplenomegaly. Laboratory tests revealed HGB 97 g/l, WBC 4.5x10^9/l, PLT 183x10^9/l and LDH 382 U/l. The liver and renal functions, total protein levels, a coagulation test, and CSF, CMV and EBV tests were normal. A BM smear and BMB were normal. The CT scan revealed hilar liver lymphadenopathy and mild splenomegaly. The child was finally diagnosed as having primary ileocecal region MALToma stage IIIA with aaIPI 2 and ECOG 1. He received CHOP [cyclophosphamide (CTX), adriamycin (ADM), vincristine (VCR) and prednisone], Hyper-CVAD/MA [CTX, VCR, ADM, dexamethasone (Dex) / methotrexate (MTX), cytarabine (Ara-C), methylprednison] and BFM 90 regimens, as well as intrathecal MTX, Ara-C and Dex five times. He achieved CR. One year after the end of treatment, the tumor recurred with periaortic lymph node enlargement and splenomegaly. He was treated with four cycles of the CHOP regimen again and achieved CR_2. Four months later, he underwent APBSCT. The conditioning regimen was BEAM. The child tolerated the transplant process fairly well. Fifty months after APBSCT and more than 6 years after diagnosis he is still in good health.

Case 3. A 10-year-old male was admitted to our hospital in May 2011 with a 10-day history of right inguinal mass. Physical examination revealed a 7x8-cm right inguinal lymph node and no hepatosplenomegaly. Laboratory tests revealed HGB 128 g/l, WBC 7.1x10^9/l, PLT 548x10^9/l and LDH 346 U/l. The liver and renal functions, total protein levels, a coagulation test, and CSF, CMV and EBV tests were normal. A BM smear revealed 84.5% lymphoma cells. BMB revealed involvement by lymphoma cells. FCM of BM revealed 83.8% abnormal cells. Cells were positive for CD19, CD20, $CD79_a$, CD10, CD34, cTdT and Ki67, and negative for CD3, CD4, CD8, CD23, CD56, FMC7, κ, λ, IgD and IgM. The chromosome was 46 xy. The CT scan revealed right parailiac vessels, and retroperitoneal and right inguinal lymphadenopathy. An inguinal lymph node biopsy was performed. Pathological examination

revealed intermediate mononuclear cells containing nucleoli. IHC staining revealed positivity for CD10, CD19, CD20, $CD79_a$, TdT and bcL2, Ki67 >75%$^+$, and negativity for CD3, CD5, CD21, bcL6, CD45RO, cyclin D1 and MUM-1. The diagnosis was B-LBL/leukemia. The patient received BFM 90 and Hyper-CVAD/MA regimens, as well as intrathecal MTX, Ara-C and Dex eight times. He achieved CR. The child was finally diagnosed as having B-cell acute lymphoblastic leukemia/lymphoma, standard risk group. Five months later, he underwent APBSCT. He tolerated the conditioning regimen (BuCy plus rituximab). Subsequent to APBSCT, the patient was alive without evidence of disease after 35 months of follow-up.

Case 4. A 15-year-old boy was admitted to our hospital in January 2013 with a 20-day history of palpitation, shortness of breath and a right cervical mass. Physical examination revealed a 3x5-cm right cervical lymph node, with breath sound diminished in the bilateral lower lobe zones. Laboratory tests revealed HGB 153 g/l, WBC $6.1x10^9$/l, PLT $132x10^9$/l and LDH 177 U/l. The liver and renal functions, total protein levels, a coagulation test, and CSF, CMV and EBV tests were normal. A BM smear revealed 4.5% lymphoid cells. BMB was normal. The CT scan revealed bilateral cervical lymph node enlargement, a 4.2x9.6-cm mediastinal tumor, hilar lymphadenopathy, pericardial and bilateral pleural effusion, and splenomegaly. A lymph node biopsy was performed. Pathological examination revealed lymphoid cell infiltration. IHC staining revealed positivity for CD3, CD7, CD99 and TdT, Ki67 80%$^+$, and negativity for CD20, $CD79_a$ and MPO. The diagnosis was T-cell lymphoblastic lymphoma (T-LBL). The patient received thoracentesis. Bloody pleural fluid (2,500 ml) was drained. The child was treated with the BFM 90 regimen, intrathecal MTX, Ara-C and Dex eight times. The assessment of PET-CT was CR. The child was finally diagnosed as having T-LBL stage IVA with aaIPI 2, ECOG 2, and high-risk group. Three months later, he underwent APBSCT. The conditioning regimen was BEAM. Following transplantation, he is now well and in CCR after 15 months of follow-up.

Discussion

Hematopoietic stem cell transplantation has demonstrated excellent treatment results in children and adolescents with NHL in the last few decades. Won *et al* reported on 33 pediatric patients who underwent APBSCT in 11 institutes. The study revealed that APBSCT was an effective treatment for certain patients with refractory or recurrent NHL. Statistically significant unfavorable factors were observed only in children with a histological subtype of lymphoblastic lymphoma and non-CR status at the time of transplantation (9). Our data support these results. Prior to APBSCT, nine patients with CR and CR_u were alive. One patient with non-CR status succumbed to progressive disease. Wojcik *et al* revealed that the survival rate is higher in children with NHL due to their chemosensitivity. The conditioning regimen BEAM was demonstrated to be a safe procedure (13). In our study, two patients experienced relapse. They achieved CR_2 as they were chemotherapy-sensitive. They survived following APBSCT. Nine patients received the BEAM regimen with better efficacy and less toxicity. The transplantation-related complications were minimal. Niemann and Thomas reported the case an 8-year-old girl with B-LBL of a vertebral body (L_3) isolated lytic lesion. She was treated symptomatically and did well for 4 months until she developed increasingly severe backache (14). Our case 1, with similar B-LBL of the vertebral body (T_6) underwent treatment and achieved CR. The result revealed that patients should not give up treatment.

Since the introduction of monoclonal anti-CD20 antibody, rituximab has provided a notable impact on the outcome of treatment for B-cell lymphoma. Attias and Weitzman reviewed all published data on rituximab therapy in patients of B-LBL, BL and large B-cell lymphoma. Among their cases, three children achieved CR following combined chemotherapy and rituximab. One of them was treated with autologous hematopoietic stem cell transplantation plus rituximab. The side effects of rituximab were usually mild. This result suggests that rituximab may play a significant role in treating B-NHL in children as well as in adults with high-grade B-NHL (5). Alasaad and Barr described the case of an 8-year-old child with T-cell rich B-NHL who was treated with chemotherapy. He had his first relapse one month after the cessation of treatment. He achieved a second remission following salvage chemotherapy and allo-HSCT. Eight months after the transplant he relapsed again and this time CR was achieved through the use of rituximab as a single agent. Eight years after rituximab therapy the patient remains in good health (6). In our series, the addition of rituximab to the BEAM and BuCy regimens was used for four patients, and was demonstrated to be a safe agent. It has not yet been determined whether the use of rituximab contributes to the disappearance of the minimal residual disease and increases the response rate. In our study there were a small number of patients and the follow-up time was short. Further research into a larger number of cases is therefore required to further explore this issue.

References

1. Angotti R, Marini M, Giannotti G, *et al*: Gastric Burkitt's lymphoma in a child: A rare case. Oncol Lett 4: 802-804, 2012.
2. Gross TG, Hale GA, He W, *et al*: Hematopoietic stem cell transplantation for refractory or recurrent non-Hodgkin lymphoma in children and adolescents. Biol Blood Marrow Transplant 16: 223-230, 2010.
3. Sun X, Zhen Z, Zhu J, *et al*: Outcomes of modified NHL-BFM-90 protocol for children and adolescents with lymphoblastic lymphoma. Zhonghua Xue Ye Xue Za Zhi 35: 1083-1089, 2014 (In Chinese).
4. Huang W, Zou D, Sui W, *et al*: Clinical outcome of autologous stem cell transplantation as first-line treatment in 30 patients with high risk lymphoblastic lymphoma. Zhonghua Xue Ye Xue Za Zhi 35: 332-336, 2014 (In Chinese).
5. Attias D and Weitzman S: The efficacy of rituximab in high-grade pediatric B-cell lymphoma/leukemia: a review of available evidence. Curr Opin Pediatr 20: 17-22, 2008.
6. Alasaad T and Barr R: Successful treatment of multiply relapsed lymphoma with rituximab as a single agent. Pediatr Blood Cancer 55: 356-358, 2010.
7. Harris RE, Termuhlen AM, Smith LM, *et al*: Autologous peripheral blood stem cell transplantation in children with refractory or relapsed lymphoma: results of Children's Oncology Group Study A5962. Biol Blood Marrow Transplant 17: 294-258, 2011.
8. Fujita N, Mori T, Mitsui T, *et al*: The role of hematopoietic stem cell transplantation with relapsed or primary refractory childhood B-cell non-Hodgkin lymphoma and mature B-cell leukemia: a retrospective analysis of enrolled cases in Japan. Pediatr Blood Cancer 51: 188-192, 2008.

9. Won SC, Han JW, Kwon SY, *et al*: Autologous peripheral blood stem cell transplantation in children with non-Hodgkin's lymphoma: A report from the Korean society of pediatric hematology-oncology. Ann Hematol 85: 787-794, 2008.
10. Giulino-Roth L, Ricafort R, Scaradavou A, *et al*: Ten-year follow-up of pediatric patients with non-Hodgkin lymphoma treated with allogeneic or autologous stem cell transplantation. Pediatr Blood Cancer 60: 2018-2024, 2013.
11. Andion M, Molina B, Gonzalez-Vicent M, *et al*: High-dose busulfan and cyclophosphamide as a conditioning regimen for autologous peripheral blood stem cell transplantation in childhood non-Hodgkin lymphoma patients: A long-term follow-up study. J Pediatr Hematol Oncol 33: e89-e91, 2011.
12. Swerdlow SH, Campo E, Harris NL, *et al* (eds): WHO Classification of Tumors of Haematopoietic and Lymphoid Tissues. IARC Press, Lyon, 2008.
13. Wojcik KB, Kowalczyk JR, Chybicka A, *et al*: Autologous stem cell transplantation in children with non-Hodgkin lymphomas. Przegl Lek 61 Suppl 2: 53-56, 2004 (In Polish).
14. Niemann TH and Thomas PA: Primary lymphoma of bone: diagnosis by fine-needle aspiration biopsy in a pediatric patient. Diagn Cytopathol 12: 165-167, 1995.