Leukemia Research, Vol. 26(2), 215-219, 2002

Case Report:

Acute T-lymphoblastic leukemia relapsed with the character of myeloid/NK cell precursor phenotype: A case report.

Shigeo Hashimoto, Ken Toba, Sadao Aoki, Junjiro Tsuchiyama, Nobuhiro Tsukada, Hidenobu Takahashi, Masuhiro Takahashi and Yoshifusa Aizawa.

First Department of Internal Medicine, Niigata University School of Medicine, Niigata, Japan
Corresponding author: Ken Toba, M.D. First Department of Internal Medicine, Niigata University School of Medicine, Asahimachi-dori 1, Niigata City, 951-8510, Japan.
Tel.: (Japan) (0)25/227/8125, Fax: (Japan) (0)25/227/0774, e-mail: tobaken@med.niigata-u.ac.jp

Key words: T-ALL, myeloid/NK cell precursor acute leukemia, Pro-T lymphoblast, acute leukemia, CD3 epsilon, CD56.

Abstract The leukemic lymphoblasts of a patient expressed CD7, CD13, CD33, CD34, HLA-DR and cytoplasmic CD3ε. He was diagnosed with ALL, and successfully treated with a conventional chemotherapy for ALL. The disease relapsed 3 times, and the character of the cells gradually altered, i.e. CD56 expression increased and CD13, CD7 and cCD3ε decreased. The phenotype of the relapsed ALL was, therefore, compatible with M/NK-AL. Some of M/NK-AL may be closely related with T/myeloid-biphenotypic pro-T blasts, and both types of AL may develop a tendency to express myeloid antigens, and they may belong to the category of immature T lymphoid precursors.

List of abbreviations: AL, acute leukemia; ALL, acute lymphoblastic leukemia; AML, acute myelogeneous leukemia; M/NK-AL, myeloid/NK cell precursor acute leukemia; LBL, lymphoblastic lymphoma; CR, complete remission; BMT, bone marrow transplantation; NK, natural killer; DC, dendritic cell; MPO, myeloperoxidase.

Introduction

The differential diagnosis of AL is of critical importance to clinical hematologists. Most cases with AML or ALL have been successfully treated with daily intensive combined chemotherapy for 5 to 7 days, and sometimes 10 to 14 days for resistant cases, or weekly moderate combination chemotherapy, respectively. CR of AML is achieved after bone marrow aplasia, and the CR of ALL is mostly achieved without bone marrow aplasia. In contrast, the differential diagnosis of AL is complex and difficult in blasts with similar character to hematopoietic stem cells. Efficient diagnosis depends on the knowledge of hierarchy of the stem cells and lymphoid and myeloid precursors, and on the technology to discriminate between these immature cells.

The FAB classification of AL has added an entity of peroxidase-negative immature AML (M0) [1]. ALs with simultaneous expression of the definite myeloid and lymphoid phenotypes are classified as biphenotypic ALL [2, 3]. A peroxidase-negative AL with myeloid phenotype on the surface and cytoplasmic expression of CD3 may be misdiagnosed as AML (MO) when the cytoplasmic expression of CD3, CD5 and CD22 is not analyzed. Moreover, there remain at least two lymphoid cell lineages left to be further studied, other than the T and B lymphocytes, i.e. NK cells and lymphoid DCs. Information about native NK, NK precursors and their transformed cells is steadily being reported, while there is almost no information about AL and LBL transformed from lymphoid DC precursors. The M/NK-AL was proposed by Suzuki et al. [4, 5]. Is the obscure entity of AL myeloid or lymphoid, or really a transformation of the NK precursors?

The blastic cells from a patient with T/myeloid biphenotypic ALL showed only myeloid markers on the surface despite the expression of cytoplasmic CD3ε. The phenotype of the cells gradually altered each time the disease relapsed; the final phenotype being compatible to the so called M/NK-AL. Some of the entity of the M/NK-AL is therefore closely related to the pro T-ALL with myeloid phenotype, and both types of ALL may belong to a spectrum of immature T lymphoid precursor ALs.

Case report

A 22-year-old Japanese man was admitted to our hospital for marked leukocytosis in September, 1995. His white cell count was 66,420 /μl, and 97% of the cells were morphologically peroxidase-negative lymphoblasts. The surface phenotype of the blasts was HLA-DR, CD7, CD13, CD15, CD33, CD34, CD38, CD45-positive, and glycophorin A, CD1a, CD2, CD3, CD4, CD8, CD10, CD14, CD19, CD20, CD21, CD22, CD25, CD36, CD41, CD56, CD122-negative. The cytoplasmic antigen expression was analyzed [6], and the cells were MPO, cytoplasmic CD22 (cCD22)-negative, and cCD3ε-positive (Fig 1). The cell cycle analysis of the blasts was also analyzed using 7AAD/PY [7, 8]. The percentages of G1a, G1b, S, G2M, and the levels of the nucleic acid index (NI) and the coefficient of variations of the RNA in the S-phase (SCV) were 75.1%, 13.8%, 7.9%, 3.3%, 1.83 and 11.5, respectively. The NI value was intermediate between Type I (lymphoid precursors, NI < 1.7) and Type 0 (lineage-negative precursors, 2.0 < NI < 2.3) [9]. Chromosomal analysis was performed on the bone marrow which showed normal karyotype in all the metaphases studied. The rearrangements of the TCR and immunoglobulin genes were evaluated in the bone marrow sample by Southern blotting analysis. The sample showed rearranged band for TCR/Cβ gene. The rearranged band was weaker than that of germline of the TCR/Cβ gene, and the reason was unclear. The sample did not show rearranged bands for JH, Jκ or Jλ of immunoglobulin genes. Analysis for bcr/abl was not performed. He was diagnosed with pro-T ALL, and successfully treated with a DVP regimen as conventional chemotherapy for ALL; weekly intravenous injections of daunorubicine and vincristine, and daily oral administrations of predonisolone. CR was achieved soon after in October, 1995. After several courses of consolidation and intensification chemotherapy, he received a BMT from an unrelated healthy donor in July, 1996.

The clinical course after the BMT was fair for 8 months, but the disease relapsed in March, 1997. The leukocyte count at that time was 11,940 /μl, and the percentages of the blastic cells in the blood and bone marrow were 89% and 92%, respectively. The phenotypic analysis of the cells showed a decreased CD15-positive percentage, a decreased intensity of CD7 and cCD3ε-antigen expression, and the appearance of CD56-positive blasts. The percentages of G1a, G1b, S, G2M, and the levels of the NI and the SCV were 85.0%, 7.0%, 6.0%, 2.1%, 2.01 and 14.9, respectively. The NI value belonged to Type 0. He was then treated again with DVP chemotherapy, and the 2nd CR was achieved in May, 1997.

In December, 1998, he suffered from headache and lower limb pain, and the cerebrospinal fluid showed 880 /μl of involved blasts. The observation of the bone marrow aspirate at that time maintained CR. The phenotypic analysis of the blasts showed the disappearance of CD15 and CD7, and full expression of CD56, and cCD3ε was almost negative. He was treated with several intrathecal injections of methotrexate, and then he received spinal radiotherapy.

The disease relapsed again in the bone marrow in August, 1999. He did not accept further chemotherapy, and the blast percentage in the blood increased to 93%, to 7,730 /μl leukocytes in November, 1999. The blasts at that time showed a low degree of expression of CD7, CD13 and cCD3ε, and almost all of the cells were CD56-positive. He died of a massive gastrointestinal hemorrhage in January, 2000.

Discussion

The European Group for the Immunological Characterization of Leukemia (EGIL) has proposed that T-lineage ALL is defined as blasts with the expression of cytoplasmic or membrane CD3, and that AML expresses two or more antigens of MPO, CD13, CD33, CD65 and/or CD117 [2]. A scoring system for the diagnosis of biphenotypic leukemia has also been proposed, and this type of leukemia tends to express CD7, CD34, CD38 and HLA-DR. The system has regarded cytoplasmic CD3 and MPO as important markers for lineage definition, and the expression of CD13, CD33 and/or CD15 less so as myeloid markers, even though the expression of CD15 helps the diagnosis of CD13/CD33-negative AML [10]. The phenotype of the blastic cells at onset was therefore compatible with biphenotypic T-ALL, and the patient was successfully treated with a conventional chemotherapy for ALL.

M/NK-AL, argued to be the abnormal counterpart of common precursor for myeloid and NK cell lineages [4, 11], expressed CD7, CD33, CD34 and CD56 on the surface, and showed dim expression of CD3 in the cytoplasm by flow cytometry and/or Northern blotting. The immunophenotype of the blasts at the relapses

was compatible to M/NK-AL. Although the blastic cells showed normal karyotype, and therefore there was no evidence that the blastic cells at onset and the cells at the relapses arose from the same clone, both sets of cells supposed to have arisen from the same clone because the expression of cCD3ε in the blasts gradually decreased, and phenotype of the pathological cells were basically very similar except CD56, they were therefore likely to be subclones of the original leukemia, but not one or more clones arisen from second leukemia at each relapse after BMT.

It is not clear whether NK cells arise from the supposed common myeloid/NK cell precursor or from the lymphoid precursor. CD3-/CD56+ NK cells were induced from CD34+/CD33+/CD56- progenitors in vitro [12], and CD34+/CD33+ progenitors do not mean strictly myeloid-committed progenitors, but they may include B- and T-lymphocyte progenitors. Although it is not known at which stage of maturation of the NK cell lineage the CD56 molecules express, CD56 is negative at the immature stage of NK progenitors. In contrast, T-lymphocytes, B-lymphocytes, NK-cells and lymphoid DC arise from CD34+/CD10+, lineage marker-negative common lymphoid precursors [13, 14]. NK cells, therefore, may arise not from the supposed CD56-positive common myeloid/NK cell precursor, but from CD56-negative common lymphoid precursor. If so, the alteration of the immunophenotype in this case may explain the close relationship between the progenitor-T cells and a part of the so called M/NK-AL that expresses cytoplasmic CD3ε, and may mean that supposed M/NK-AL is not the abnormal counterpart of common precursor for myeloid and NK cell lineages, and moreover, the expression of CD56 in this part of so-called M/NK-AL does not mean it is an NK-marker.

CD3-positive or CD56-positive mature lymphocytes include heterogeneous subsets, and therefore the clarification and definition of the subsets are confusing. CD3-positive cells include thymic- and extrathymic-T cells, or CD56-negative and positive-T (NK-like T) cells. The majority of CD56-positive T cells, that are of extrathymic origin, show granular lymphocyte morphology and NK-activity to a low degree [15, 16]. In other words, the wide spectrum of killer lymphocytes includes cells supposed to be primitive to sophisticated killers, HLA-non restricted and restricted cells, i.e., CD3+/CD8+/CD56- activated killer T cells, CD3+/CD56+ NK-like T cells, and CD3-/CD56+ natural killers. Most of the cells express CD3ε, perforin and granzyme B in the cytoplasm, and therefore, whatever the precursors of the cells are, the precursors may show similar phenotype. NK cells in fetal liver, cord blood and decidua express cCD3ε [17, 18]. It is also unclear whether the immature cells named progenitor-T and precursor-T are strictly the progenitors for thymic-T or the common progenitors of this vast spectrum of non-B lymphocytes. To understand the hematopoiesis of the cells, it is important to study the in vivo and in vitro maturation of the precursors, as well as to analyze the malignant counterparts of the cells in detail, such as in AL and LBL.

References

1. Bennett JM, Catowsky D, Danier MT, Flandrin G, Galton DA, Gralnick HR, Sultan C. Proposal for the recognition of minimally differentiated acute myeloid leukemia (AML-M0). Br J Haematol 1990; 78: 325 - 9.
2. Bene MC, Castoldi G, Knapp W, Ludwig WD, Matutes E, Orfao A, van't Veer MB. Proposals for the immunological classification of acute leukemias. Leukemia 1995; 9: 1783 - 6.
3. Rothe G, Schmitz G. Consensus protocol for the flow cytometric immunophenotyping of hematopoietic malignancies. Working Group on Flow Cytometry and Image Analysis. Leukemia 1996; 10: 877 - 95.
4. Suzuki R, Yamamoto K, Seto M, Kagami Y, Ogura M, Yatabe Y, Suchi T, Kodera Y, Morishima Y, Takahashi T, Saito H, Ueda R, Nakamura S. CD7+ and CD56+ myeloid/natural killer cell precursor acute leukemia: a distinct hematolymphoid disease entity. Blood 1997; 90: 2417 - 28.
5. Murase T. Suzuki R. Tashiro K. Morishima Y. Nakamura S. Blast crisis of chronic myelogenous leukemia exhibiting immunophenotypic features of a myeloid/natural killer cell precursor. Int J Hematol 1999; 69: 89 - 91.
6. Toba K, Koike T, Shibata A, Hashimoto S, Takahashi M, Masuko M, Azegami T, Takahashi H, Aizawa Y. Novel technique for the direct flow cytofluorometric analysis of human basophils in unseparated blood and bone marrow, and the characterization of phenotype and peroxidase of human basophils.

Cytometry 1999; 35: 249 – 59.
7. Toba K, Winton EF, Koike T, Shibata A. Simultaneous three-color analysis of the surface phenotype and DNA-RNA quantitation using 7-Amino-actinomycin-D and Pyronin Y. J Immunol Methods 1995; 182: 193 - 207.
8. Toba K, Kishi K, Koike T, Winton EF, Takahashi H, Nagai K, Maruyama S, Furukawa T, Hashimoto S, Masuko M, Uesugi Y, Kuroha T, Tshukada N, Shibata A. Profile of cell cycle in hematopoietic malignancy by DNA/RNA quantitation using 7AAD/PY. J Exp Hematol 1996; 24: 894 - 901.
9. Toba K, Koike T, Watanabe K, Fuse I, Takahashi M, Hashimoto S, Takahashi H, Abe T, Yano T, Shibazaki Y, Itoh H, Aizawa Y. Cell kinetic study of normal human bone marrow hematopoiesis and acute leukemia using 7AAD/PY. Eur J Haematol 2000; 64: 10 – 21.
10. Khalidi HS, Medeiros J, Chang KL, Brynes RK, Slovak ML, Arber DA. The immunophenotype of adult acute myeloid leukemia. High frequency of lymphoid antigen expression and comparison of immunophenotype, French-American-British classification, and karyotypic abnormalities. Am J Clin Pathol 1998; 109: 211 – 20.
11. Suzuki R, Nakamura S. Malignancies of natural killer (NK) precursor: myeloid/NK cell precursor acute leukemia and blastic NK cell lymphoma/leukemia. Leuk Res 1999; 23: 615 – 24.
12. Shibuya A, Nagayoshi K, Nakamura K, Nakauchi H. Lymphokine requirement for the generation of natural killer cells from CD34+ hematopoietic progenitor cells. Blood 1995; 85: 3538 – 46.
13. Galy A, Travis M, Cen D, Chen B. Human T, B, natural killer and dendritic cells arise from a common bone marrow progenitor cell subset. Immunity 1995; 3: 459 – 73.
14. Galy A, Christpherson I, Ferlazzo G, Liu G, Spits H, Georgopoulos K. Distinct signals control the hematopoiesis of lymphoid-related dendritic cells. Blood 2000; 95: 128 – 37.
15. Ortaldo JR, Winker-Pickett RT, Yagita H, Young HA. Comparative studies of CD3- and CD3+CD56+ cells: examination of morphology, function, T cell receptor rearrangement, and pore-forming protein expression. Cellular Immunol 1991; 136: 486 – 95.
16. Takii Y, Hashimoto S, Iiai T, Watanabe H, Hatakeyama K, Abo T. Increase in proportion of granulated CD56+ T cells in patients with malignancy. Clin Exp Immunol 1994; 97: 522 – 7.
17. Phillips JH, Hori T, Nagler A, Bhat N, Spitz H, Lanier LL. Ontogeny of human natural killer (NK) cell: Fetal NK cells mediate cytolytic function and express cytoplasmic CD3ε, δ proteins. J Exp Med 1992; 175: 1055 – 66.
18. King A, Gardnerm L, Sharkeym A, Loke YW. Expression of CD3ε, CD3ζ and RAG-1/RAG-2 in decidual CD56+ NK cells. Cellular Immunol 1998; 183: 99 – 105.

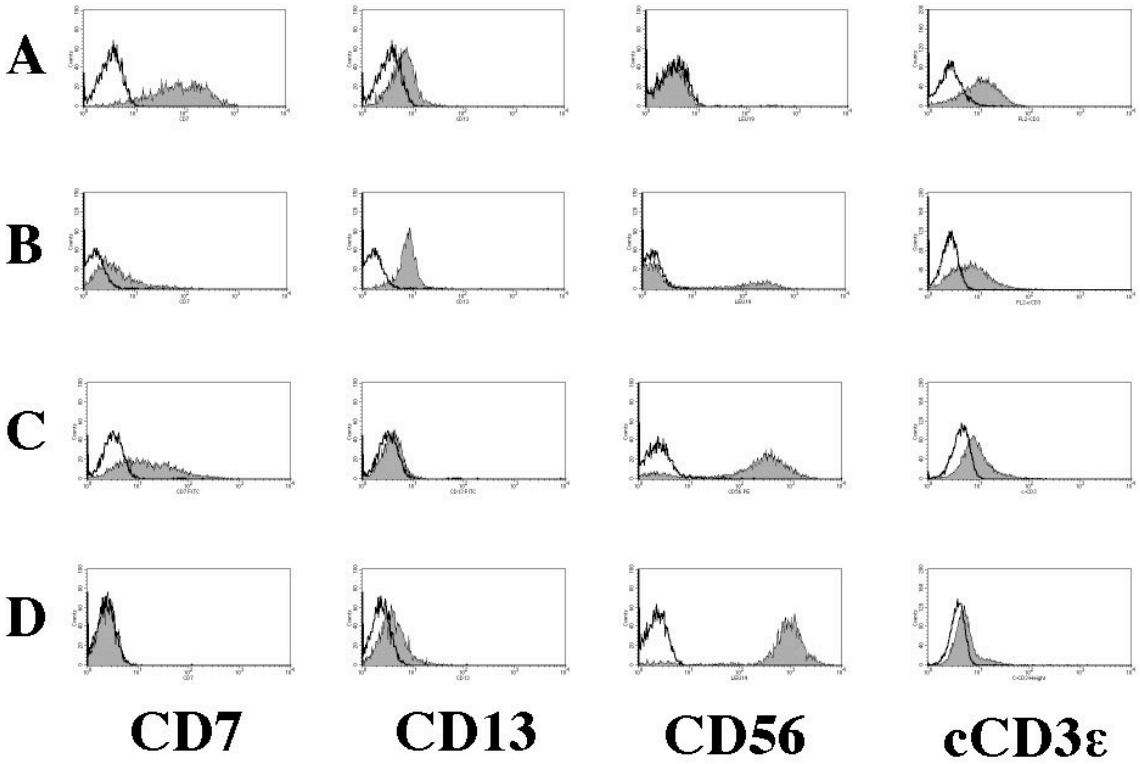

Fig.1 The phenotype alteration of the blastic cells. Surface expressions of CD7, CD13, CD56, and cytoplasmic expression of CD3ε are shown. A, onset, peripheral blood (PB) in September, 1995; B, 1st bone marrow relapse, bone marrow (BM) in March, 1997; C, 2nd bone marrow relapse, PB in November, 1999; and D, meningeal relapse, liquor in December, 1998.

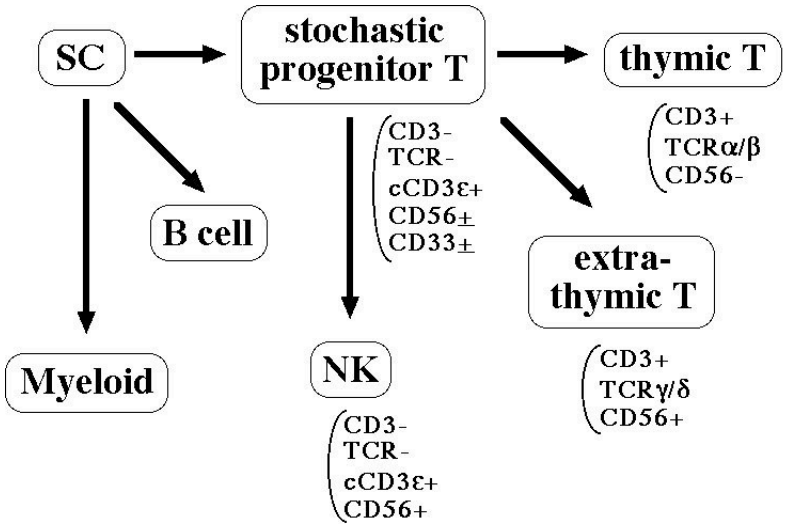

Fig.2 Possible scheme of cCD3ε-positive progenitor. SC, hematopoietic stem cell; cCD3, intracytoplasmic CD3. Note that this scheme is simplified by disregarding minorities as TCRα/β extrathymic T, CD56-negative extrathymic T, CD2+/CD3-/CD56-/CD57+ strange granular lymphocytes, etc.