shorter PCR product consisted of the first 2 exons of *RCSD1* fused similarly to exon 4 of *ABL1* (Figure 2B). These two different fusion transcripts were likely caused by alternative splicing of the fusion gene. The predicted oncogenic product of *RCSD1-ABL1* is in-frame and encodes the tyrosine kinase domain of ABL.

Most previously described fusion genes involving *ABL1* (*BCR, ETV6, EML1, NUP214*) fuse with exon 2 of *ABL1*.[3-5] However, similar to *RCSD1-ABL1*, a novel fusion partner (*SFPQ*) was recently reported to fuse to exon 4 of *ABL1* in a pre-B-ALL patient with a t(1;9)(p34;q34) translocation.[6] The fusion protein resulting from *ABL1* exon 4 fusion lacks the amino acids W127-K183 to form intact SH2 domain.[6] Murine experiments have shown that BCR-ABL constructs with inactivated SH2 domain are able to induce B-ALL-like disease,[7] which further supports the central role of *RCSD1-ABL1* in the development of leukemia in our patient. However, additional mechanisms may also exist, as the *RCSD1* gene encodes a protein kinase substrate CapZIP which interacts with the actin capping protein CapZ. This may influence the cytoskeleton regulation and/or migration of the cells.[8]

In Ph$^+$ALL dasatinib is a promising novel treatment option.[1] Our case report describes that TKIs may have clinical efficacy in other types of B-ALL as well. Furthermore, the clinical activity of dasatinib was recently described in a T-ALL patient carrying a *NUP214-ABL1* fusion.[9] Although these fusion genes may occur rarely, the therapeutic implications warrant that they be searched for in acute leukemia. The screen for such uncommon, but clinically significant fusion transcripts should be included in multiplex PCR panels used in the routine diagnostic work-up. Our data further underline the importance of dividing hematologic malignancies into molecularly defined and clinically meaningful disease entities, allowing for a rational and effective selection of optimal treatment modalities for each patient.[10]

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Funding: this work was supported by the Finnish special governmental subsidy for health sciences, research and training, by the Finnish Cancer Societies, Emil Aaltonen Foundation, Academy of Finland, Finnish Medical Foundation, Biomedicum Helsinki Foundation and KA Johansson foundation.

Acknowledgments: the authors would like to thank the personnel at the Department of Pathology, Laboratory of Molecular Genetics, Helsinki University Central Hospital and Tampere University Hospital for their help with patient sample analysis.

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Citation: Mustjoki S, Hernesniemi S, Rauhala A, Kähkönen M, Almqvist A, Lundán T, and Porkka K. A novel dasatinib-sensitive RCSD1-ABL1 fusion transcript in chemotherapy-refractory adult pre-B lymphoblastic leukemia with t(1;9)(q24;q34). Haematologica 2009;94:1469-1471. doi: 10.3324/haematol.2009.008094

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Advanced Philadelphia chromosome positive acute lymphoblastic leukemia patients relapsed after treatment with tyrosine-kinase inhibitors: successful response to clofarabine and cyclophosphamide

Philadelphia chromosome positive acute lymphoblastic leukemia (Ph$^+$ ALL) includes at least one-quarter of adults with ALL. Treatment with tyrosine-kinase inhibitors (TKIs), with or without chemotherapy, today represents the most appealing management both in terms of complete remission (CR) and disease-free survival (DFS), and towards providing eligible patients with a *bridge* to hemopoietic stem cell transplantation (HSCT).[1-4] However, relapsed Ph$^+$ ALL is still regarded as an almost incurable disease. Clofarabine, a second generation deoxyadenosine analog, has demonstrated significant activity in children and adults with refractory lymphoid and myeloid leukemia in early clinical trials.[5] To improve its single-agent antileukemic activity different clofarabine combinations are being studied.[6] With the clofarabine-

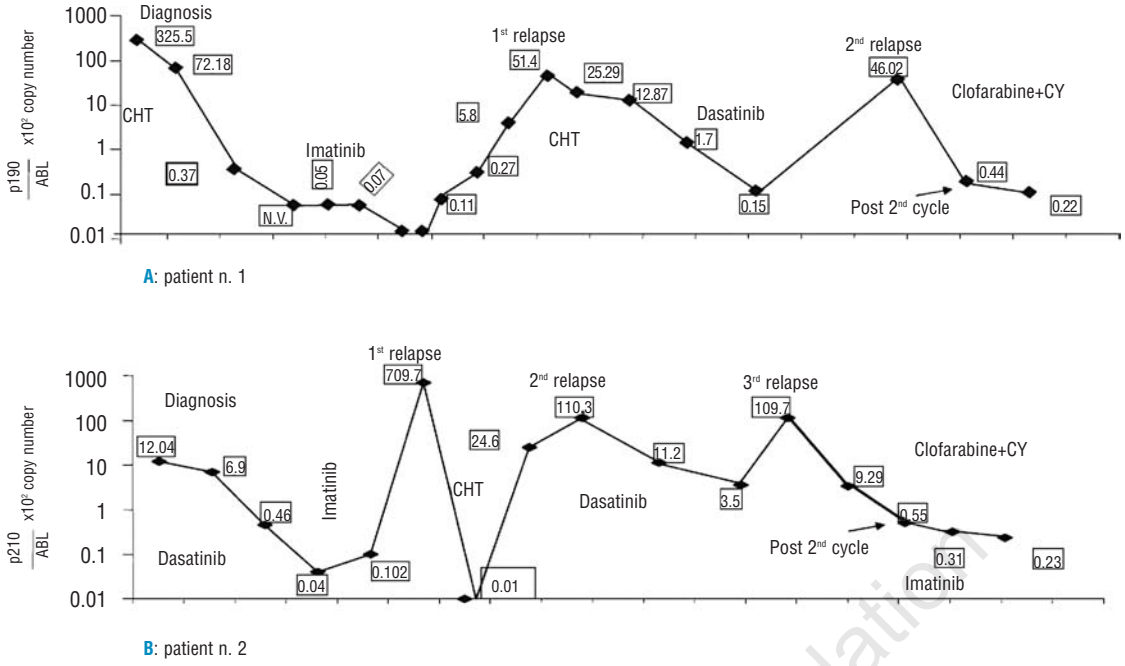

Figure 1. Molecular monitoring of BCR-ABL. The graphics show the molecular monitoring of BCR-ABL levels by quantitative reverse-transcriptase polymerase-chain-reaction (Q-RT-PCR) in the 2 patients. Values obtained were normalized with respect to the number of ABL transcripts and expressed as a percentage of ABL (BCR-ABL/ABLx10²); patient transcript ratios were converted to a logarithmic (base 10) scale of reduction from individual baseline ratio BCR-ABL/ABL assayed at diagnosis. (A) BCR-ABL p190 levels in patient n. 1 during follow-up. After the consolidation cycle, the combination of clofarabine + cyclophosphamide produced a molecular response with a 2-log reduction of the BCR-ABL transcript, which was maintained until the time of transplant. (B) BCR-ABL p210 transcript levels in patient n. 2. A 1-log reduction of BCR-ABL was observed after the consolidation cycle of therapy with clofarabine + cyclophosphamide. The transcript levels decreased further at the start of maintenance treatment with imatinib and remained at low levels at the time of the report.

cyclophosphamide combination, two phase I studies testing different doses and schedules have been reported in relapsed adult ALL; several dose-limiting toxicities were observed.[7,8]

We report our experience on 2 adult female patients with Ph⁺ ALL in advanced phase and refractory to treatment with chemotherapy and TKIs. The first patient was a 57-year old female, diagnosed in August 2005 with BCR-ABL p190 ALL, and treated according to the GIMEMA LAL 2000 protocol,[9] which provided a maintenance therapy with imatinib 600 mg/day. The patient, 17 months from the first CR (after 15 months of imatinib), suffered a first relapse and molecular analysis revealed the presence of the ABL point mutation Y253H. Salvage treatment with vincristine, daunorubicin and prednisone (PDN) allowed a second CR to be obtained; consolidation therapy was planned according to the GIMEMA LAL 0288 protocol.[10] The patient started maintenance treatment with dasatinib (70 mg BID) and monthly intrathecal methotrexate. After five months (eight months from the second CR), a second relapse occurred.

The second patient was a 59-year old female, diagnosed in February 2007 with BCR-ABL p210 ALL and treated according to the GIMEMA LAL 1205 protocol, consisting of front-line therapy with dasatinib and PDN alone.[3] After achieving CR, the patient continued therapy with imatinib (600 mg/day) up to the first relapse four months later (five months from CR). Molecular analysis revealed the presence of the E255K-ABL point mutation. She received salvage treatment and consolidation as the other patient. Three months after the second CR, a second relapse occurred and dasatinib (70 mg BID) was re-administered; the patient achieved a new CR, but a third relapse occurred four months later (one month after the third CR) and the molecular analysis showed a T315I-ABL point mutation. The patient received clofarabine 40 mg/m² by a one hour intravenous (iv) infusion followed by cyclophosphamide 400 mg/m² by a one hour iv infusion, from day 1 to 5, as induction phase. G-CSF was initiated at the onset of neutropenia and continued up to a neutrophil count ≥1.0×10⁹/L. Prophylactic PDN (10 mg/m²/daily) was given to prevent potential systemic inflammatory response. Allopurinol 300 mg/day was administered to prevent the occurrence and complications of tumor lysis–induced hyperuricemia. Antimicrobial, viral and fungal prophylaxis consisting of trimetoprim-sulfametoxazole and ciprofloxacin, acyclovir and itraconazole was administered. Consolidation with clofarabine and cyclophosphamide was given, at the same doses as the first cycle, for three days with the same supportive treatment. The trial was approved by the local ethics committee and carried out in accordance with the Declaration of Helsinki. All analyses of peripheral blood (PB) and BM are part of the investigatory work-up for all adult ALL cases entering the GIMEMA trials.[11] The presence of ABL point mutations was assayed as previously described.[12] The first patient exhibited hematologic recovery at day +20 and a BM aspirate performed at day +21 confirmed the CR. Moreover, after consolidation, a molecular response consisting of a 2-log reduction of the BCR-ABL transcript (Figure 1A) was also seen. One month later, the patient underwent an HSCT from a mis-

matched unrelated donor and died of respiratory distress syndrome. The second patient exhibited hematologic recovery on day +16 and the BM aspirate performed on day +17 confirmed the CR. Moreover, after the consolidation, a molecular response consisting of a 1-log reduction of the BCR-ABL transcript (Figure 1B) and absence of the T315I-ABL mutation was also evident. The patient was unsuitable for an HSCT and imatinib (600 mg/day) was re-started as maintenance therapy, taking into account both the disappearance of the ABL point mutation and the possibility of utilizing a second-generation TKI as further salvage therapy. After five months from clofarabine-cyclophosphamide treatment, she is still in CR with a low number of BCR-ABL transcript copies (Figure 1B) and absence of ABL mutations. The clofarabine-cyclophosphamide combination was very well tolerated. No patient suffered from nausea, vomiting or dose-limiting toxicities. Possibly in view of the rapid hematologic recovery and strict antimicrobial prophylaxis, neither patient developed infections.

Clofarabine followed by cyclophosphamide seems to be a very promising salvage treatment for adult patients with advanced ALL, even in the presence of adverse prognostic factors like the BCR-ABL rearrangement, relapse after TKI administration and presence of the T315I mutation. Clinical trials on large series of patients will conclusively clarify the efficacy and safety of this combination.

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Key words: adult Ph positive acute lymphoblastic leukemia, clofarabine, tyrosine-kinase inhibitors.

Funding: CAF is an employee of Genzyme (Genzyme S.r.l, Italy). Other authors indicate that they have no potential conflicts of interest.

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Citation: Vitale A, Grammatico A, Capria S, Fiocchi C, Foà R, Meloni G. Advanced Philadelphia chromosome positive acute lymphoblastic leukemia patients relapsed after treatment with tyrosine-kinase inhibitors: successful response to clofarabine and cyclophosphamide. Haematologica 2009;94:1471-1473. doi: 10.3324/haematol.2009.011718

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Transient loss of consciousness in pediatric recipients of dimethylsulfoxide (DMSO)-cryopreserved peripheral blood stem cells independent of morphine co-medication

Toxicity related to the infusion of dimethylsulfoxide-cryopreserved peripheral blood stem cells (DMSO-PBSC) manifests mostly as cardiovascular side effects. Neurotoxicity[1] including transient global amnesia,[2,3] seizures,[4,5] and stroke[6,7] has been reported as a rare complication primarily in adults. In children, data[8] are sparse. In light of a recent report implicating morphine co-medication as a major contributing factor,[8] we evaluated retrospectively our own data base, including all infusions of DMSO-PBSC applied in our pediatric center between January 1st 2002 and December 31st 2008. We report on 2 incidences of transient loss of consciousness following 131 infusions of DMSO-PBSC.

Case 1. A 17-year old female suffering from recurrent medulloblastoma was admitted for prolonged pancytopenia due to topotecan maintenance therapy. Throughout her medical history, this patient had received intensive therapy, including high-dose chemotherapy and autologous peripheral blood stem cell transplantation, craniospinal irradiation, as well as CNS-directed therapy, namely intrathecal chemotherapy and radioimmunotherapy (Table 1). Thirty minutes after receiving a cryopreserved autologous stem cell boost, the patient initially complained of bilateral loss of vision. Fifteen minutes later, she became unconscious (Glasgow coma scale 3/15). The patient's cardiorespiratory condition was stable. During transport to emergency CT, she developed one short episode of clonic seizure restricted to both arms, which