Imatinib-induced long-term remission in a relapsed *RCSD1-ABL1*-positive acute lymphoblastic leukemia

The treatment of patients with *BCR-ABL1*-positive chronic myeloid leukemia and acute lymphoblastic leukemia (ALL) with tyrosine kinase inhibitors (TKIs) has significantly improved their overall survival.[1-3] Several reports confirm that this type of treatment may be similarly effective also in cases with other types of kinase or cytokine receptor signaling activating fusion genes, particularly those involving the *ABL1, ABL2, JAK2, PDGFRB, EPOR, CRLF2* and *CSF1R* genes.[1-7] However, follow-up times are generally short, with treatment still ongoing at the time of publication.[6] It, therefore, remains an open question whether continuous long-term remissions can be achieved and sustained, even after discontinuation of TKI treatment. Here we present the case of a young adolescent with a relapsed B-cell precursor (BCP) ALL and a rare *RCSD1-ABL1* gene fusion, in whom transitory treatment with imatinib induced a continuing remission that to date lasts eleven years.

In April 2001, a 15-year old boy was admitted to the Pediatric Hematology/Oncology Department of the Medical University in Graz because of increasing pallor and deteriorating physical performance. His white blood cell count (WBC) was 68.7x10^9/L with 71% blasts, platelets 93x10^9/L and hemoglobin 44 g/L. The 95% blasts in the bone marrow (BM) were CD10, CD19, cyCD22, CD79a, CD34, TdT and HLA-DR positive and negative for myeloid and T-cell markers. Cytogenetic analysis revealed an abnormal clone with a 46,XY,t(1;9)(q31?;q34), which was shown by fluores-

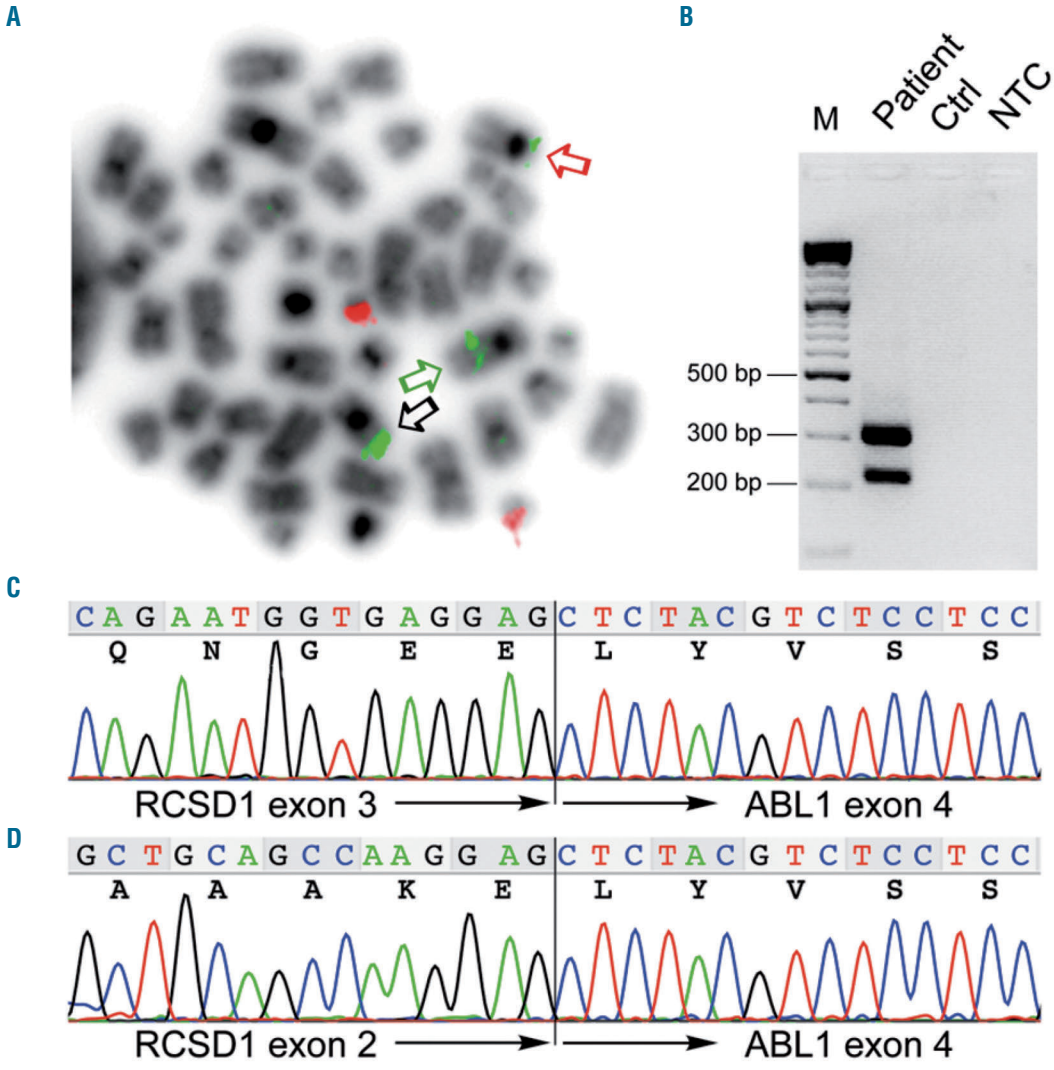

Figure 1. Detection of the *RCSD1-ABL1* rearrangement. (A) Fluorescence *in situ* hybridization (FISH) of a metaphase obtained from the bone marrow at relapse using the BCR/ABL Dual Color, Dual Fusion Translocation Probe (Oncor) showing two signals for *BCR* (red signals) and three signals for *ABL1* (green signals). Black arrow indicates the *in situ* signal on the normal chromosome 9, the green and the red arrows the *ABL1* signals on the der(9) and der(1) chromosomes, respectively. (B) RT-PCR of *RCSD1-ABL1* fusion transcripts using primers RCSD1ex1_2-F1 (5'-CCTGAAGGACATGGAGGAAAGACC-3') spanning exons 1 and 2 of *RSCD1* and ABL1ex4-R1 (5' CTGGATAATGGAGCGTGGTGATG-3') located in exon 4 of *ABL1* showing two distinct amplification products. (C and D) Sequence chromatograms corresponding to the two fusion transcript variants detected by RT-PCR. In-frame fusions of (C) *RCSD1* exon 3 to *ABL1* exon 4 and (D) alternatively spliced *RCSD1-ABL1* lacking *RCSD1* exon 3. M: molecular weight marker; Ctrl: control, normal cDNA; NTC: non-template control.

Table 1. Clinical and hematologic characteristics of reported patients with *RCSD1-ABL1*+ ALL.

Case	Phenotype	Year of diagnosis/ report	Sex	Age (y)	WBC x10^9/L	PB blasts (%)	BM blasts (%)	Treatment	Relapse (m)	Survival (m)	Cytogenetics	Ref.
A	Biphenotypic leukemia	2004	M	15	122 23$_R$	95 95$_R$	NA NA$_R$	Chemotherapy	10	10†	t(1;9)(q23.3~q25;q34)	13,15
1	BCP-ALL	2003	M	11	6$_R$	47$_R$	92$_R$	Chemotherapy + HSCT	11	97	t(1;9)(q24;q34)	4,8,12,13
2	BCP-ALL	2009	M	40	24	34	80	Chemotherapy + dasatinib + HSCT; Chemotherapy + dasatinib/imatinib$_R$	16	66	t(1;9)(q24;q34)	4,10,12,13
3	BCP-ALL	2010	M	31	146	90	NA	Chemotherapy + imatinib/dasatinib	No CR	6.5†	t(1;9)(q23;q34)	13
4	BCP-ALL	2010	F	18	110	87	92	NA	NA	NA	t(1;9)(q24;q34)	11,12
5	BCP-ALL	2011	F	15	348	NA	NA	Chemotherapy + HSCT	3/33/75	84†	t(1;9)(q24;q34)	4,12
6	BCP-ALL	2012	M	15	48	NA	NA	NA	NA	NA		5
7	BCP-ALL	2013	M	18	470	52	58	No treatment compliance	No CR	12	t(1;9)(q24;q34)	12
8	BCP-ALL	2014	M	6	108	NA	NA	Chemotherapy + imatinib	–	1	t(1;9)(q24;q34)	6
9	BCP-ALL	2012	F	26	26	84	86	Chemotherapy + dasatinib + HSCT; Chemotherapy$_{R1}$; ponatinib + HSCT$_{R2}$; ponatinib$_{R3}$	15/19/24	25†	t(1;9)(q24;q34)	14
10	BCP-ALL	2001	M	15 18$_R$	69 24$_R$	71 64$_R$	95 80$_R$	Chemotherapy; chemotherapy + CNS irradiation + imatinib$_R$	39	163	t(1;9)(q31?;q34) t(1;9)(q25?;q34)$_R$	Present work

(A) RCSD1-ABL1 not assessed; (1) bone marrow transplantation (BMT) at 31 months (m); (2) refractory disease after induction, + dasatinib: complete remission (CR) after 1 m, continued until hematopoietic stem cell transplantation (HSCT) at 4 m/relapse (R) at 16 m, chemotherapy + dasatinib: CR, then dasatinib monotherapy, switched to imatinib due to immunological side effects; (3) partial response to chemotherapy and dexamethasone (DXM) + imatinib/dasatinib/progressive disease (PD) after 3 m: partial response to chemotherapy and DXM + dasatinib/PD at 6 m: no response, death; (4) no further data reported; (5) R 3, 33 and 75 m after diagnosis, BMT at 4, 35 and 84 m; (6) no further data reported; (7) no treatment compliance; (8) poor response to induction, imatinib: residual disease 2% after 1 m; (9) chemotherapy + dasatinib: 0.2% residual disease, continued until HSCT at 5 m/R1 at 15 m: CR1 under chemotherapy/R2 at 19 m: CR under ponatinib monotherapy, continued until HSCT2 at 22 m/R3 at 24 m: ponatinib monotherapy: response, severe graft-versus-host disease (GvHD) and death due to septic shock; (10) chemotherapy + imatinib 2 m after relapse: sustained CR after 1 m: y: years; WBC: white blood cells; PB: peripheral blood; BM: bone marrow; TKI: tyrosine kinase inhibitor; M: male; F: female; NA: no data available; †: death; m: months.

cence *in situ* hybridization (FISH) using a BCR/ABL Dual Color, Dual Fusion Translocation Probe (Oncor) to disrupt the *ABL1* gene. The description of *RCSD1* as the *ABL1* fusion partner in an identical translocation in 2007 facilitated the subsequent identification of the respective *RCSD-ABL1* fusion gene by RT-PCR and sequencing also in our case (Figure 1).[8]

The patient was stratified into the intermediate risk group of the AIEOP-BFM ALL 2000 study protocol and treated accordingly. The day 15 BM aspirate still contained 62% blasts. Morphological remission was achieved on day 33, although immunoglobulin gene rearrangement-based minimal residual disease (MRD) was still detectable on days 33 and 77 at levels of 10^{-3} and 10^{-4}, respectively (Figure 2A). After 39 months, the patient experienced a BM relapse with a WBC of 24x10^9/L and 64% blast cells in the peripheral blood. Therefore, further treatment was initiated according to the ALL-REZ BFM 2002 protocol. Due to therapy refractory disease, this regimen was complemented with daily oral doses of 400 mg imatinib on day 64 of relapse treatment. This decision was driven not only by the fact that the patient was resistant to conventional therapy, but also by the presence of an *ABL1* gene fusion (although at that time the partner gene had not yet been identified). Morphological remission was achieved on day 68, whereas MRD levels remained positive until day 79 but became negative by day 109 (Figure 2B). Imatinib was given during the entire course of relapse chemotherapy and was continued following the patient's explicit request and the treating physicians' discretion for another 69 months as mono-therapy before it was discontinued on the basis of a joint decision and with the patient's approval. He has now been without any treatment for more than 36 months and remains in remission almost 15 years after disease onset. Although this response pattern clearly indicates that conventional chemotherapy was adequate to reduce the disease burden, it is not known whether it would have been sufficient to cure the patient on its own.

In addition to *BCR*, eight other *ABL1* fusion partners are currently known in B-cell precursor ALL, namely *ETV6, ZMIZ1, NUP214, FOXP1, SNX2, RANBP2, SFPQ* and *RCSD1* and four, *NUP214, INPP5D (SHIP1), EML1* and *SEPT9* in T-cell precursor ALL.[2,4-6,8-14] TKI treatment has already shown very promising results in a consider-

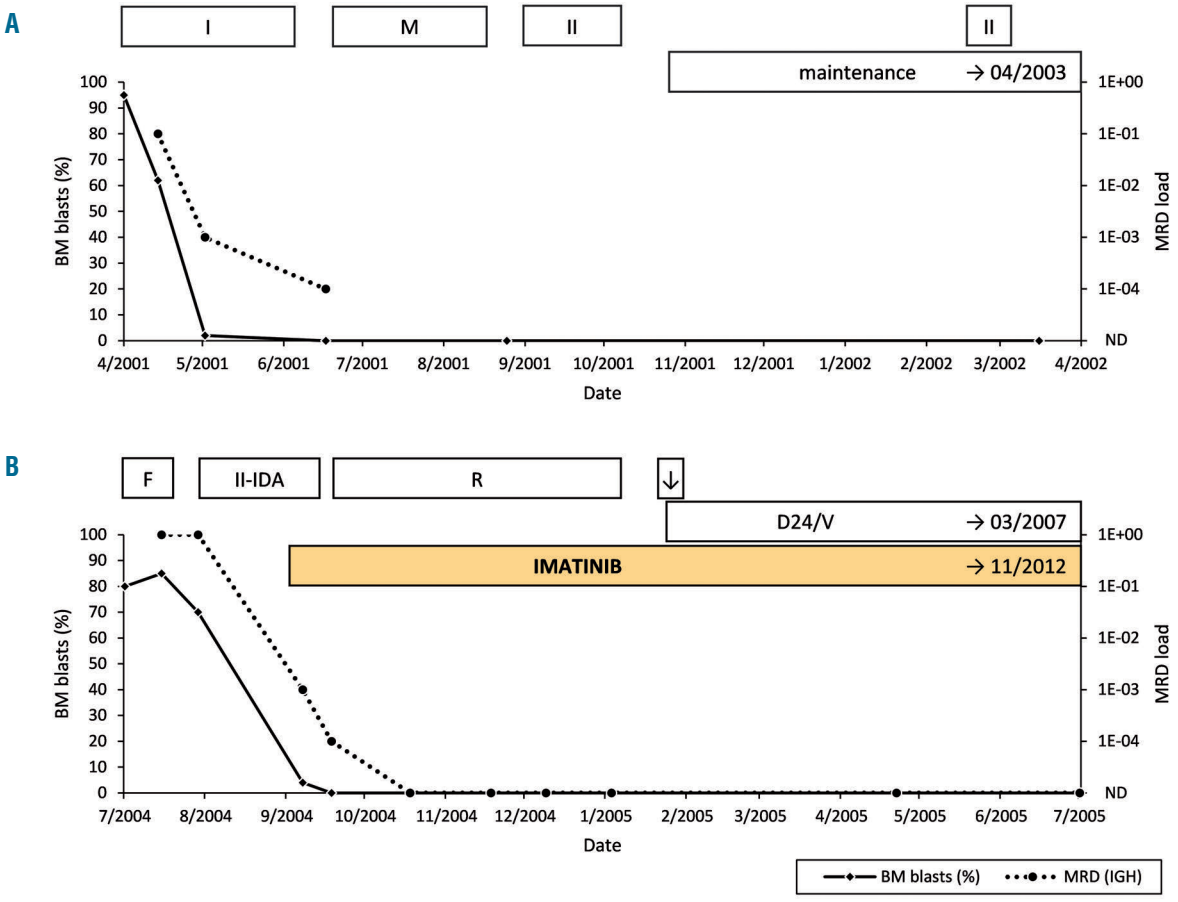

Figure 2. Course of initial disease and relapse. (A) Initial disease: course of leukemic blasts detected in bone marrow (%) and minimal residual disease (MRD) load (immunoglobulin heavy chain locus; QR: 1E-04) under treatment according to the ALL-BFM-2000 study protocol [Prot. IA-P+ and IB (I); Prot. M (M); Prot. II (II); maintenance therapy]; maintenance therapy was continued until April 2003 [BM: complete remission (CR)]. (B) Relapse: course of leukemic blasts detected in bone marrow (%) and minimal residual disease (MRD) load (immunoglobulin heavy chain locus; QR: 1E-04) under treatment according to the ALL-REZ-BFM-2002 study protocol – S_2A [Prot. F1 and F2 (F); Prot. II-IDA (II-IDA); Prot. R1 and R2 (R); CNS irradiation (↓); maintenance therapy (D24/V)]; maintenance therapy was continued until March 2007; imatinib treatment was continued until November 2012. Further bone marrow assessment: complete remission in April 2006, 2007 and 2008, MRD not detectable in April 2008.

able number of patients with such kinase-activating gene fusions,[1,2,4-7,9,10] although they may become refractory to this therapy in a similar fashion to *BCR-ABL1*-positive CML and ALL. Refractoriness may be due to various molecular mechanisms, such as ABL1 kinase domain mutations, additional genetic lesions, as well as signaling pathway alterations.[1-4,12-14] Resistance can, at least in part, be overcome with 2nd- and 3rd-generation TKIs, such as dasatinib, nilotinib, bosutinib and ponatinib, which do not only affect different conformational states of the respective fusion proteins, but also a broader spectrum of tyrosine kinases.[1-4,6,14]

Ten cases with B-cell precursor ALL and a molecular genetically verified *RCSD1-ABL1* fusion gene have been reported so far, including the patient described herein (Table 1).[4-6,8,10-14] An identical translocation was also found in an *ABL1*-positive biphenotypic ALL case in which, however, the partner gene has not been identifed.[15] Four of these patients were treated with TKIs and all of them achieved at least a partial remission (Table 1).[6,10,13,14] To the best of our knowledge, however, historically our patient is not only the first *RCSD1-ABL*-positive case, but also the first one ever with a variant *ABL1* fusion who has been rescued with imatinib treatment and cured despite cessation of therapy.

Our instructive case underscores the importance of identifying these particular genetic lesions, as they may become highly relevant for appropriate treatment decisions. The choice of TKI is generally based on the specific gene rearrangement and the associated constitutively activated kinase. As exemplified in our *ABL1*-positive case, even the knowledge of the particular kinase alone may already be enough to select the most appropriate first-line TKI (in this case imatinib). In fact, this drug is not only effective in *BCR-ABL1*-positive cases, but may perhaps also be effective in a similar fashion in those with other *ABL1* kinase-activating gene fusions. The treatment with 2nd- and 3rd-generation TKIs may thus be restricted only to cases that are either primarily refractory, become resistant or relapse. Owing to the low number and the consequential general lack of experience with cases that harbor such rarer types of tyrosine-kinase activating fusion genes, the advantages and disadvantages of when and if at all an apparently successful TKI treatment should be stopped must still be carefully weighed up and, therefore, the final decision can only be made on an individual basis.

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