

Letter to the Editor

Primary resistance to imatinib in Fip1-like 1–platelet-derived growth factor receptor α —positive eosinophilic leukemia

To the Editor:

The Fip1-like 1 (FIP1L1)–platelet-derived growth factor receptor α (PDGFRA) gene fusion is a common cause of chronic eosinophilic leukemia (CEL), and patients with this particular subgroup of CEL respond to low-dose imatinib therapy.^{1,2} However, some patients may develop imatinib resistance because of

acquired T674I mutation.² Here, we describe a FIP1L1–PDGFRA–positive patient with CEL with primary resistance to imatinib. A 59-year-old white man has had idiopathic hypereosinophilic syndrome since 1989. Since then, the patient developed endocarditis fibroblastica, pulmonal hypertension, multifocal cortical ischemia, polyneuropathy (restless legs syndrome), splenomegaly, renal insufficiency, and a restrictive pulmonary disease. An investigation of his bone marrow in 1997 confirmed the diagnosis hypereosinophilic syndrome with no evidence for cytogenetic abnormalities or increased numbers of blasts. Over

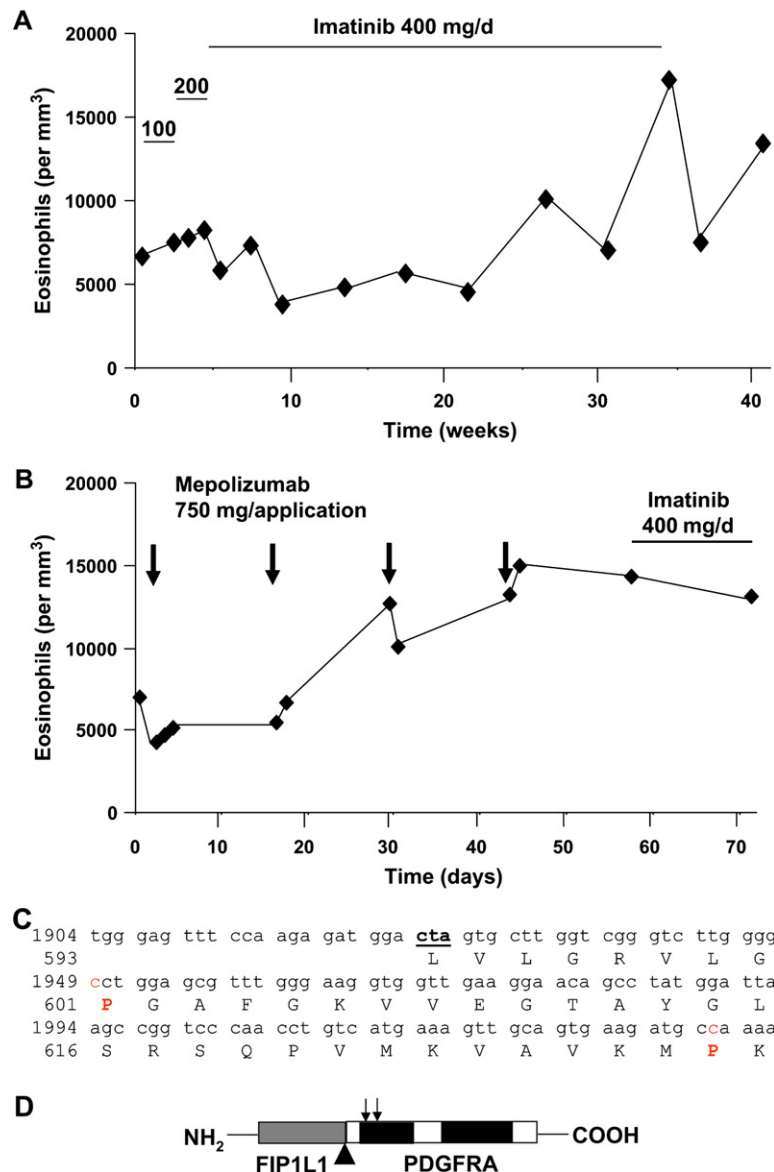


FIG 1. Primary imatinib resistance in FIP1L1–PDGFRA–positive CEL associated with 2 mutations in the kinase domain of PDGFRA. **A**, Effect of imatinib on blood eosinophil numbers. **B**, Effect of 4 mepolizumab applications and subsequent imatinib treatment on blood eosinophil numbers. **C**, Nucleotide and amino acid sequences of PDGFRA (M22734). The first indicated amino acid represents the start of the kinase domain. Mutations are marked in red. **D**, Simplified scheme of the FIP1L1–PDGFRA protein. The position of the breakpoint is indicated by an arrowhead. The approximate locations of the 2 novel mutations, S601P and L629P, in the first part of the split kinase domain (black boxes) of PDGFRA are indicated by arrows.

the years, the patient was treated with various amounts of prednisolone alone or in combination with IFN- α (1990-1993), hydroxyurea (1994 and 2000), or imatinib mesilate (2003; Fig 1, A). The peripheral blood eosinophil numbers were not reduced by these therapies (range, $3\text{--}18 \times 10^3/\text{mm}^3$), and the patient experienced disease progression. Leukocyte numbers were usually elevated (range, $12\text{--}22 \times 10^3/\text{mm}^3$).

The patient presented in September 2003, complaining about itchy skin rash, fatigue, dyspnea and cough, and depressive episodes. Immunophenotyping of his blood lymphocytes revealed a CD4/CD8 ratio of 0.8 with no evidence for a T-cell clone.³ Both CD4 and CD8 T cells demonstrated increased expression of the activation markers HLA-DR and CD95. The blood eosinophils expressed high levels of CD25, suggesting an IL-5-driven mechanism.⁴ Indeed, IL-5 in serum was 23.3 pg/mL (normal, <2.0 pg/mL). Increased levels of serum IL-5 were confirmed in 2 subsequent measurements. By using RT-PCR, we identified the presence of the FIP1L1-PDGFR α gene fusion.² We also identified FIP1L1-PDGFR α protein at the expected size by immunoblotting in purified eosinophil populations of this patient (data not shown). These data suggested that the patient had CEL associated with increased T-cell activation and elevated IL-5 serum levels. Previously published work demonstrated that patients with the FIP1L1-PDGFR α gene fusion respond to imatinib mesilate.^{2,5} However, FIP1L1-PDGFR α -positive patients with CEL may also respond to anti-IL-5 antibody treatment,⁶ perhaps because the FIP1L1-PDGFR α gene fusion alone is not sufficient and may require IL-5 to induce CEL.⁷

To understand whether the eosinophil differentiation process can be stopped by neutralizing IL-5, we administered 4 intravenous injections of an anti-IL-5 antibody (mepolizumab, 750 mg each) with intervals of 2 weeks.⁸ The patient did not respond with a detectable decrease of his eosinophil count as a consequence of this treatment (Fig 1, B), suggesting that the neutralization of IL-5 by mepolizumab was not sufficient to stop the eosinophil differentiation process. The antibody treatment had also no influence on the relative distribution of T subpopulations and B cells or the expression of markers indicating their activation.

Although single imatinib treatment had no effect (Fig 1, A), we thought a combination of mepolizumab and imatinib mesilate might be able to reduce eosinophil numbers. Therefore, we treated the patient with 400 mg imatinib daily, starting 2 weeks after the last mepolizumab infusion. At this time, mepolizumab still exhibits antieosinophil activity in hypereosinophilic patients.⁸ Again, no effect of imatinib on eosinophil numbers or symptoms was seen (Fig 1, B). The patient was subsequently substituted with 25 mg hydrocortisone. He died of heart failure in 2006.

Because imatinib resistance might be caused by mutations, we analyzed the primary structure of the entire FIP1L1-PDGFR α fusion gene. To this end, the complete FIP1L1-PDGFR α cDNA was amplified by RT-PCR and sequenced. We identified 2 mutations in PDGFR α sequence (accession no. M22734: T1949C and T2034C), resulting in 2 amino acid changes within the kinase domain: S601P and L629P (Fig 1, C and D). No homologous mutations have been described in the BCR-ABL fusion

oncoprotein causing imatinib resistance in chronic myeloid leukemia.⁹ Therefore, the exact molecular mechanisms through which these newly identified mutations likely cause imatinib resistance remain to be investigated. It should be noted that no mRNA was available before the patient received the drug for the first time. Therefore, the possibility exists that the imatinib treatment may have influenced the mutation. Nevertheless, mutations within the kinase domain of oncoproteins are common causes of drug resistance.¹⁰

Taken together, patients with CEL with the FIP1L1-PDGFR α gene fusion may be resistant to imatinib mesilate because of mutations within the tyrosine kinase domain of the chimeric protein, and increased IL-5 levels in blood do not predict an antieosinophil response to anti-IL-5 antibody therapy. The identification of novel drug-resistant variants may help to develop the next generation of target-directed compounds for CEL and other leukemias.

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