The efficacy of imatinib mesylate in patients with *FIP1L1-PDGFRα*-positive hypereosinophilic syndrome. Results of a multicenter prospective study

Michele Baccarani, Daniela Cilloni, Michela Rondoni, Emanuela Ottaviani, Francesca Messa, Serena Merante, Mario Tiribelli, Francesco Buccisano, Nicoletta Testoni, Enrico Gottardi, Antonio de Vivo, Emilia Giugliano, Ilaria Iacobucci, Stefania Paolini, Simona Soverini, Gianantonio Rosti, Francesca Rancati, Cinzia Astolfi, Fabrizio Pane, Giuseppe Saglio, Giovanni Martinelli

From Department of Hematology-Oncology "L. and A. Seràgnoli", University of Bologna, and S.Orsola-Malpighi University Hospital, Bologna (MB, MR, EO, NT, AdV, II, SP, SS, GR, GM); Department of Clinical and Biological Sciences, University of Turin at Orbassano, and S.Luigi Gonzaga Hospital, Orbassano (DC,FM, EG, EG, GS); Division of Hematology, IRCCS S. Matteo, Pavia (SM); Division of Hematology, Udine University and General Hospital, Udine (MT); Division of Hematology, University Tor Vergata, Rome (FB); CEINGE Advanced Biotechnologies and Department of Biochemistry and Medical Biotechnology, University Federico II, Naples (CA, FP).

Acknowledgments: Paolo Ricci (Bologna); Pier Ferruccio Ballerini (Conegliano); Filippo Gherlinzoni, Cristina Danesin and Cristina Tecchia (Treviso); Sergio Morandi (Cremona); Ercole De Biasi (Castelfranco Veneto); Paolo Vivaldi (Trento); Barbara Izzo (Naples); Pier Paolo Fattori (Rimini); Eliana Zuffa and Barbara Giannini (Ravenna); and Antonio Spadea (Rome) referred and cared for several patients. Their co-operation is gratefully acknowledged.

Funding: the study was supported by COFIN 2003 and Fondazione del Monte di Bologna e Ravenna grants.

Manuscript received February 23, 2007.
Manuscript accepted June 27, 2007.

Correspondence:
Michele Baccarani, Department of Hematology-Oncology "L. and A. Seràgnoli", S.Orsola-Malpighi University Hospital, Via Massarenti, 9 40138 Bologna, Italy.
E-mail: michele.baccarani@unibo.it

ABSTRACT

Background and Objectives

The hypereosinophilic syndrome (HES) may be associated with the fusion of the platelet derived growth factor receptor α (*PDGFRα*) gene with the *FIP1L1* gene in chromosome 4 coding for a constitutively activated PDGFRα tyrosine kinase. These cases with *FIP1L1-PDGFRα* rearrangement have been reported to be very sensitive to the tyrosine kinase inhibitor imatinib mesylate.

Design and Methods

A prospective multicenter study of idiopathic or primary HES was established in 2001 (Study Protocol Registration no. NCT 0027 6929). One hundred and ninety-six patients were screened, of whom 72 where identified as having idiopathic or primary HES and 63 were treated with imatinib 100 to 400 mg daily.

Results

Twenty-seven male patients carried the *FIP1L1-PDGFRα* rearrangement. All 27 achieved a complete hematologic remission (CHR) and became negative for the fusion transcripts according to reverse transcriptase polymerase chain reaction (RT-PCR) analysis. With a median follow-up of 25 months (15-60 months) all 27 patients remain in CHR and RT-PCR negative, and continue treatment at a dose of 100 to 400 mg daily. In three patients imatinib treatment was discontinued for few months, the fusion transcript became rapidly detectable, and then again undetectable upon treatment reassumption. Thirty-six patients did not carry the rearrangement; of these, five (14%) achieved a CHR, which was lost in all cases after 1 to 15 months.

Interpretation and Conclusions

All patients meeting the criteria for idiopathic or primary HES should be screened for the *FIP1L1-PDGFRα* rearrangement. For all patients with this rearrangement, chronic imatinib treatment at doses as low as 100 mg daily ensures complete and durable responses.

Key words: eosinophils, hypereosinophilic syndrome, FIP1L1-PDGFRα, tyrosine kinase, imatinib.

Haematologica 2007; 92:1173-1179. DOI: 10.3324/haematol.11420

©2007 Ferrata Storti Foundation

©Ferrata Storti Foundation

Thee term hypereosinophilic syndrome (HES) covers a relatively wide range of conditions in which eosinophilia is apparently primary or idiopathic (i.e. is not associated with other recognizable disease entities), is consistent (more than 1.5×10^9 eosinophils per liter), is durable (for more than 6 months), and may be associated with symptoms and signs of organ or tissue involvement and dysfunction.[1] So far HES treatment, including corticosteroids, hydroxyurea, and interferon-α, has been symptomatic or palliative, and of limited success.[1,2] The causes and the nature of such cases of primary eosinophilia remained obscure until it was shown that in some cases of eosinophilia associated with a myelodysplastic syndrome the platelet derived growth factor receptor beta (*PDGFRβ*) was constitutively activated upon fusion with different partner genes, so providing a cytokine independent signal for cell proliferation.[3-8] These rare cases of MDS are exquisitely responsive to the protein tyrosine kinase inhibitor imatinib mesylate, which is able to inhibit the kinase activity of PDGFR, at nanomolar concentrations.[9,10] Soon after, imatinib was reported to be effective also in the treatment of other patients with primary eosinophilia,[11,15] and it was rapidly found that in these patients an interstitial deletion in chromosome 4 led to the formation of a novel gene by fusion of the *FIP1L1* gene with the *PDGFRα* gene, coding for a tyrosine kinase that is a constitutively activated form of PDGFRα,[16-18] and can also be inhibited by imatinib at nanomolar concentrations.[10] The cases of HES that are based on this peculiar molecular abnormality may be renamed as cases of chronic eosinophilic leukemia (CEL).[2,19] In a short time, several such cases were identified and were shown to be very sensitive to imatinib.[17,30,31] There is fairly good evidence that imatinib is likely to be the treatment of choice of *FIP1L1-PDGFRα* rearranged HES/CEL, but the number of cases is small, and the long-term results of treatment are unknown. We report here on 27 patients with the *FIP1L1-PDGFRα* rearrangement who have been treated with imatinib for 15 to 60 months (median 25 months).

Design and Methods

Study and treatment protocol

A prospective study and treatment protocol of primary eosinophilia was approved by the Ethics Committee of the S. Orsola-Malpighi University Hospital in 2001 and subsequently by the Ethics Committees of the of other 19 Italian academic institutions, in which the study was run from December 2001 to present (study protocol registration number NCT00276929). The study protocol required that patients with primary eosinophilia (more than 1.5×10^9 eosinophils/L for more than 6 months) were screened for cytogenetic and molecular studies, including the *FIP1L1-PDGFRα* rearrangement, and were eligible for an experimental treatment with imatinib. Prior treatment was always discontinued for at least 15 days with the exception of corticosteroids, which were maintained at the same dose until a response to imatinib was shown and confirmed for 15 days. Imatinib was provided free-of-charge by Novartis Pharma (Origgio, Italy) in 100 mg tablets and was given in a single dose, beginning with 100 mg daily for one week. Thereafter, the daily dose was increased by 100 mg each week, and was set at 400 mg from the 4th week on. Imatinib treatment was continued for a minimum of 4 weeks in case of no hematologic response (HR), or until it was felt that it was beneficial for the patient, in case of response. During the first year, the dose was adjusted for toxicity and adverse events, according to the standard criteria for dose adjustment used in the treatment of chronic myeloid leukemia.[32,33] After 1 year of treatment, the dose could be adjusted at the investigator's discretion. The patients were visited weekly for 1 month, the 12 monthly for 1 year, and every 3 months thereafter. Blood counts and differentials were performed at each visit. In patients with *FIP1L1-PDGFRα* rearrangement, the level of the transcripts was assessed by reverse transcriptase polymerase chain reaction (RT-PCR) on marrow cells, every 3 months until confirmed negativity, and then every 6 months.

Three types of response were considered, hematologic, clinic, and molecular. A complete hematologic response (CHR) required a normal white blood cell count with a total eosinophil count of less than 0.250×10^9/L for more than 4 weeks. The HR was defined as partial (PHR) if the total eosinophil count was reduced to less than 50% of baseline value but was still higher than 0.250×10^9/L. The definition of clinical response was that all the symptoms, signs and laboratory evidence of organ and tissue involvement or dysfunction disappeared completely for more than 4 weeks. A complete molecular response required negativity of the RT-PCR for the *FIP1L1-PDGFRα* transcripts on two successive tests at a 3-month interval.

Laboratory tests

Conventional cytogenetic analysis was performed on bone marrow cells that were cultured without stimulation for 48 hours. Metaphases were G-banded with Wright's stain. Chromosome abnormalities were classified according to the International System for Chromosome Nomenclature.[34]

Fluorescence *in situ* hybridization (FISH) was performed on interphase marrow cells, using a commercially available double fusion signal D-FISH BCR-ABL probe (Oncor, Appligene, Gaithesburg, MD, USA) to exclude the involvement of the *BCR* and *ABL* genes, a bacterial artificial chromosome (BAC) bk 350N15 spanning the fibroblast growth factor receptor 1 gene (*FGFR1*) (kindly provided by Dr. Negrini, University of Ferrara) for the 8p11 breakpoint. BAC probes (kindly provided by Dr. Rocchi, University of Bari) were used for the 5q33.1 platelet derived growth factor receptor beta (*PDGFRβ*)

©Ferrata Storti Foundation

(RP 11-368O19), for the 4q12 *PDGFRα* (RP 11-231C18) and for the 4q12/*CHIC2* (RP 11-3671N). BAC DNA was isolated from cultures using a standard miniprep procedure, and labeled with fluoroscein isothiocyanate or cyanine 3.5 by nick translation All FISH analyses were performed with a Nikon fluorescence microscope (Eclipse E1000, Nikon Instruments) attached to a computer-based imaging system (Genikon) equipped with a triple band-pass filter for 4,6-diamino-2-phenyl indole (DAPI), fluorescent isothiocyanate, and rhodamine.

For molecular studies, marrow mononuclear cells were obtained by Ficoll-Hypaque density gradient separation. Aliquots of 5×10^4 cells were resuspended in 600 µL GITC, and RNA was obtained following standard procedures. A qualitative RT-PCR was performed for *BCR-FGFR1*, *BCR-ABL*, and *TEL-PDGFRβ*, using standard conditions.[35] For detection of *FIP1L1-PDGFRα* fusion transcripts, a nested RT-PCR was performed according to Cools *et al.*[17] The fusion was analyzed using primers FIP1L1-F1 (5'-acctggtgctgatctttctgat) and PDGFRα-R1 (5'-tgagagcttgtttttcactgga) during the first PCR, and primers FIP1L1-F2 (5'-aaagaggatacgaatgggacttg) and PDGFRα-R2 (5'-gggaccggcttaatccatag) for the second PCR. In selected cases and for sequencing purposes, PCR products were cloned in pGEM-T-easy (Promega) and cloned products were sequenced on an ABI-PRISM 377 system (Perkin Elmer) according to manufacturer's instructions.

Table 1. Clinical and hematologic characteristics of study patients, according to *FIP1L1-PDGFRα* rearrangement.

	FIP1L1-PDGFRα rearrangement	*No FIP1L1-PDGFRα rearrangement*
No. of cases	27	36
Gender, Male/Female, no. of cases	27/0	25/11
Age, years, median (range)	50 (17-75)	58 (18-81)
Hemoglobin, g/L, median (range)	137 (94-165)	138 (84-180)
Platelet count, $\times10^9$/L, median (range)	191 (29-365)	228 (27-668)
WBC count, $\times10^9$/L, median (range)	10.7 (1.8-57.5)	12.2 (6.7-47.0)
Eosinophils %, median (range)	43 (20-85)	27 (13-38)
Eosinophil count $\times10^9$/L, median (range)	4.8 (1.6-16.5)	3.4 (1.5-34.9)
Serum creatinine ≥20 mg/L, no. of cases	1/27	1/37
Serum uric acid ≥60 mg/L, no. of cases	6/27	6/37
Serum LDH ≥460 U/mL, no. of cases	5/27	6/37
Organ or tissue involvement, no. of cases		
Lung	5/27	10/37
Spleen	5/27	1/37
Skin	0/27	5/37
Heart	2/27	2/37
Liver	1/27	1/37
Soft tissues	2/27	0/37
Waldeyer's ring	0/27	1/37
Intestine	0/27	1/37
Prior disease duration, months, median (range)	16 (6-125)	25 (6-209)
Prior treatment, % of cases	66%	66%

The major difference between positive and negative patients was gender, which was 100% male in the positive group. Positive patients tended to be slightly younger, to have a shorter disease history, less frequent lung and skin involvement but more frequent splenomegaly. Also, in positive patients the percentage of eosinophils, and total eosinophil count tended to be higher. A hemoglobin concentration and a platelet count higher than normal (more than 166 g/L and more than 450×10^9/L, respectively) was reported in two and one negative cases, respectively. Sixty-six per cent of positive and negative cases were pretreated with corticosteroids, hydroxyurea or interferon-α, or a combination of these drugs.

Results

Patients

One hundred and ninety-six patients with eosinophilia were screened; 72 were identified as having primary or idiopathic eosinophilia, and 63 of these provided consent to study procedures and experimental treatment. Preliminary data of some of these patients were reported orally at the meetings of the European Haematology Association[36] and the American Society of Hematology[37,38]. The case of one patient was already published separately.[24] The observation period of these 63 patients, from the first dose of imatinib, ranged between 15 and 60 months (median 25 months). Twenty-seven patients (43%) were found to carry the *FIP1L1-PDGFRα* rearrangement, while 36 (57%) were negative (Table 1). The main difference between patients with and without the rearrangement was gender, which was male in all 27 patients with the rearrangement vs 25 of 36 in negative cases. Other minor differences concerned age, with a median of 50 years in positive patients vs 58 years in negative ones; the total eosinophil count (median 4.8×10^9/L in positive patients vs 3.4×10^9/L in negative patients); skin involvement, which was recorded in only 5 patients without the rearrangement, and splenomegaly, which was reported in five patients with the rearrangement and in only one without. Prior duration of eosinophilia was also slightly different, the median being 16 months for patients with the rearrangement and 25 months for those without. Prior treatment, including corticosteroids, hydroxyurea or interferon-α, had been received by 66% of patients in both groups. All patients had a normal karyotype constitution with the exception of two patients without the rearrangement (45XY/45X, and 46XX+m) and one with the rearrangement (47XY+4 in 2 of 26 metaphases). All patients tested negative on FISH and RT-PCR for *BCR-ABL*, *FGFR1-BCR*, *TEL-PDGFRβ*, and *cKit* mutations.

Treatment results

Patients with the FIP1L1-PDGFRα rearrangement

All 27 patients with *FIP1L1-PDGFRα* rearrangement achieved a CHR within 1 month and all 27 patients have remained in continuous CHR until last contact, for 15^+ to 60^+ months (median 25^+ months) (Table 2). Of the 15 patients with organ involvement, 13 achieved also a complete clinical remission, since all symptoms, signs and laboratory evidence of lung (5 cases), spleen (5 cases), liver (1 case) and soft tissue (2 cases) involvement disappeared within 3 months. In the remaining two patients, who had cardiac involvement, the echocardiographic patterns were not significantly modified after 12 and 30 months of treatment. All 27 patients became RT-

©Ferrata Storti Foundation

Table 2. Hematologic response to imatinib treatment.

	Complete hematologic response	
Time on treatment	*FIP1L1-PDGFRα rearrangement (n=27)*	*No FIP1L1-PDGFRα rearrangement (n=36)*
1 month	27 (100%)	4 (11%)
3 months	27 (100%)	3 (8%)
6 months	27 (100%)	3 (8%)
12 months	27 (100%)	1 (3%)
Last contact	27 (100%)	0 –

All patients with the rearrangement achieved a complete hematologic response in less than 1 month and were on treatment and in continuous complete hematologic response at last contact, after 15 to 60 months (median 25 months). Thirteen of 15 positive patients with organ or tissue involvement also achieved a complete clinical response. Some negative patients also achieved a complete or a partial hematologic response, but the response was lost in all cases.

Table 3. Imatinib dose. As per protocol, all patients should have received 100 mg daily the first week, 200 mg daily the second week, 300 mg daily the third week, and 400 mg daily from the fourth week on, indefinitely in the case of response, or for 90 days in case of no response.

	FIP1L1-PDGFRα rearrangement	*No FIP1L1-PDGFRα rearrangement*
Daily dose, median and interval (mg)	339 (100-386)	319 (100-390)
No. of cases with a mean daily dose of 100 mg	4/27 (15%)	3/37 (8%)
No. of cases with a mean daily dose of 101-199 mg	2/27 (7%)	5/37 (13%)
No. of cases with a mean daily dose of 200-299 mg	4/27 (15%)	10/37 (27%)
No. of cases with a mean daily dose of 300-399 mg	17/27 (63%)	19/37 (52%)

All patients with the FIP1L1-PDGFRα rearrangement are still on treatment at a daily dose of 400 mg (8 patients), 300 mg (2 patients), 200 mg (10 patients) and 100 mg (7 patients). All patients who did not carry the FIP1L1-PDGFRα rearrangement are off-treatment.

PCR negative for the *FIP1L1-PDGFRα* fusion transcripts after 1 to 10 months of treatment (median 3 months). In 24 of them treatment has not been discontinued, and all these 24 patients have remained PCR-negative for 6+ to 56+ months (median 19 months). In three patients imatinib was discontinued after 12, 14 and 15 months of treatment. In all these 3 cases the *FIP1L1-PDGFRα* fusion transcripts became detectable again 4, 2, and 6 months after discontinuation of imatinib. In all three patients imatinib was reassumed at a dose of 200 mg daily, and the fusion transcripts disappeared after 2, 5, and 2 months of treatment.

The mean daily dose of imatinib ranged from 100 to 386 mg (median 339 mg). All patients were still on treatment at last contact, at a daily dose of 400 mg (8 patients), 300 mg (2 patients), 200 mg (10 patients), and 100 mg (7 patients) (Table 3).

Patients without the FIP1L1-PDGFRα rearrangement

Four of 36 patients achieved a CHR in 1 month, and another patient in 3 months, for an overall CHR rate of 14% (5/36). All five patients who achieved CHR lost the response after 1, 1, 6, 9, and 15 months of imatinib treatment. Another four patients achieved a PHR, which was lost in less than 6 months. The clinical and hematologic characteristics of responding patients could not be distinguished from those of non-responding patients, but four of the complete responders were younger (18 to 38 years old). The daily dose of imatinib was similar to that taken by the patients with the rearrangement (Table 3), but the treatment duration was much shorter, since treatment was discontinued earlier because of lack of response, disease progression or response loss. At last contact, all 36 patients without the *FIP1L1-PDGFRα* rearrangement were alive with eosinophilia.

Adverse events

The type and frequency of grade 2 and 3 adverse events are reported in Table 4. Hematologic toxicity was negligible and limited to grade 2 neutropenia in 7.9% of cases and grade 3 neutropenia in 3.2% of cases. Thrombocytopenia greater than grade 1 was never reported. Non-hematologic adverse events consisted mainly of myalgia and muscle cramps (9.5% grade 2 and 3.1% grade 3) and diarrhea (6.3% grade 2), but also skin rash, abdominal pain, edema, headache and paresthesia (Table 4). In the group with the *FIP1L1-PDGFRα* rearrangement, treatment was not abandoned in any patient, but was discontinued temporarily in three patients for three times and in four patients once. In the group without the rearrangement, treatment was abandoned in two patients because of grade 3 skin rash, and in one patient because of grade 3 abdominal pain. No clinical evidence of cardiac dysfunction was reported.

Discussion

In a prospective multicenter study of eosinophilia we screened 196 patients with this condition and identified the cause of eosinophilia in 124. We selected the remaining 72 patients with primary or idiopathic eosinophilia and a total eosinophil count of more than 1.5×10^9/L for more than 6 months, and studied and treated 63 of them with imatinib. Thirty-six of the 63 study patients also had symptoms or signs of organ or tissue involvement or dysfunction. The study patients were treated with imatinib and a CHR was achieved rapidly in 27/27 of the patients with the rearrangement and in 5/36 of those without. The response was stable over the whole observation period (15+ to 60+ months, median 25 months) in

©Ferrata Storti Foundation

Table 4. Frequency of all grade 2 and 3 adverse events. In *FIP1L1-PDGFRα* positive patients 61% of adverse events occurred during the first quarter of treatment, 23% during the second quarter and 16% after the first year of treatment.

	Grade 2	Grade 3
Neutropenia	7.9%	3.2%
Myalgia, muscle cramps	9.5%	3.1%
Diarrhoea	6.3%	–
Skin rash	1.6%	3.1%
Abdominal pain	1.6%	–
Edema, peripheral	3.1%	–
Edema, generalized	1.6%	–
Headache	1.6%	–
Paresthesia	–	1.6%

They were managed with temporary imatinib discontinuation (7 cases) or dose reduction. In FIP1L1-PDGFRα negative patients all the adverse events occurred during the first quarter of treatment and were a cause of permanent treatment discontinuation in two patients because of grade 3 skin rash, and in one patient for grade 3 abdominal pain.

Table 5. Summary of prior reports of cases of primary or idiopathic eosinophilia treated with imatinib. The reported maximum follow-up was 24 months, vs 15 to 60 months (median 25 months) in this study.

	N° CHR / N° treated with IM		
Reference	*FIP1L1-PDGFRα rearrangement*	*No FIP1L1-PDGFRα rearrangement*	*Follow-up*
Cools *et al.*, 2003[17]	3/5	5/5	8 months (max)
Pardanani *et al.*, 2003[18]	0/2	3/3	NR
Klion *et al.*, 2004[21]	0/0	7/7	18 months (max)
Musto *et al.*, 2004[26]	0/1	1/1	NR
Pardanani *et al.*, 2004[25]	2/8	10/11	NR
Rose *et al.*, 2004[23]	0/0	1/1	12 months
Vandenberghe *et al*, 2004[22]	0/1	4/4	NR
La Starza *et al.*, 2005[28]	2/5	7/7	NR
Roche-Lestienne *et al.*, 2005[27]	1/5	4/4	NR
Müller *et al.*, 2006[29]	0/1	1/1	21 months
Helbig *et al.*, 2006[31]	0/5	2/2	24 months (max)
Total	8/33 (24%)	45/46 (98%)	24 months (max)
This study	5/36 (14%)	27/27 (100%)	15-60 months (median 25 months)
Total	13/69 (19%)	72/73 (99%)	

NR: not reported.

all patients with the rearrangement, while all patients without the rearrangement lost their response in 1 to 15 months and discontinued imatinib treatment.

The high sensitivity to imatinib of HES/CEL with the *FIP1L1-PDGFRα* rearrangement has already been established. At least 47 cases had been reported, with an overall CHR rate of 95% (Table 5). Hoewever, many reports focused on the response rate and could not report on response duration. The maximum observation time after the first dose of imatinib was 24 months (Table 5). Now that the issue of the response rate has been settled, because it is increasingly clear that it is very close to 100%, the interest is focused on the quality, the stability, and the duration of the response. We found that with a median follow-up of 25 months, ranging from 15 to 60 months, the response is stable and durable in all patients carrying the rearrangement. Although molecular negativity is not equivalent to cure, it is the best available marker of the quality of the response. The achievement of molecular negativity was also reported in prior studies, but the stability and the duration of the negativity was not fully established.[18,21-23,25,29] We show that molecular negativity is durable and stable in all patients, but is dependent on treatment continuation, because molecular negativity was lost in three patients who discontinued imatinib and then regained when imatinib was resumed. Pending independent confirmation, this study suggests that the stability of the response requires continuation of imatinib treatment. This suggestion is supported by a recent update of imatinib treatment of myelodysplastic syndromes with the *PDGFRβ*-rearrangement, reporting that also in these cases the response was stable only if treatment was continued.[39] In prior studies the daily dose of imatinib ranged between 100 and 600 mg, with the majority of the patients being treated with 100 or 200 mg.[29,30] In this study the planned daily dose was 400 mg, but actually ranged between 100 and 400 mg, and the majority of the patients continue treatment at a daily dose of 100 or 200 mg. Since imatinib inhibits PDGFRα at nanomolar concentration, an imatinib dose of 100 or 200 mg is likely to be sufficient to obtain and maintain the response. Moreover, there are reports of cases in which the response was maintained with 100 mg every second day, and even once a week.[25,31] The issue of the dose is important both because the cost of the drug, which may even create serious obstacles to treatment continuation,[30] and because of the possible side effects. In these HES/CEL patients with the *FIP1L1-PDGFRα* rearrangement, imatinib was well tolerated (Table 4), and apparently even better than in patients with chronic myeloid leukemia. Hematologic toxicity was negligible at a dose of 400 mg daily, and non-hematologic toxicity was also limited, so that the overall compliance to treatment was excellent. However the chronic administration of a protein tyrosine kinase inhibitor may have effects difficult to predict. A recent report warned about cardiac toxicity,[40] although cardiac adverse events had not been of concern in large series.[41,42] Moreover, chronic imatinib treatment may affect bone turn-over, and calcium and

©Ferrata Storti Foundation

glucose metabolism.[43,44] It is therefore wise to keep the dose of imatinib at the minimum effective level and to carefully monitor the state of health of these highly selected patients. Before the *FIP1L1-PDGFRα* rearrangement was identified, several molecularly undefined HES patients were treated with imatinib and responded to this drug.[11,15] Clearly, these patients probably carried the rearrangement, but also patients without the rearrangement can respond to imatinib.[17,20-31] Previously, a documented CHR was reported in at least eight of 33 patients (24%) without the rearrangement (Table 5). The duration of the response was not reported for several of these patients. In our series, five of 36 (14%) patients without the rearrangement had a CHR, but all responders have relapsed on treatment. The bases for these responses are not clear, whether the target of imatinib was PDGFRα or other non-identified protein tyrosine kinases. From a clinical point of view, the indiscriminate use of imatinib or other protein tyrosine kinase inhibitors in cases in which a specific molecular target cannot be identified, cannot be recommended, but from a biological point of view the interest may be notable because of the possibility of discovering the role of other protein tyrosine kinases. Also of interest is the nearly absolute prevalence of male gender. Only two female patients have been shown to carry the rearrangement, so far.[22,28]

In conclusion, we have confirmed that imatinib is the treatment of choice for patients with *FIP1L1-PDGFRα*-rearranged HES/CEL, since almost all patients achieve and maintain complete hematologic, clinical and molecular remissions, chronic treatment is well tolerated, and responses are stable over time at doses as low as 100 mg daily.

Authors' contributions

MB is the author taking primary responsability for the paper: GM, GS, FP, GR and FR promoted and designed the study; MR, SM, MT, FB and SP took care of the clinical part of the study; DC, FM, NT, EG, II, EO and SS performed cytogenetic and molecular studies; MR, AdV and CA collected and analyzed the data; MB and GM wrote the report, with the contribution of DC, MR and GS.

Conflict of interest

MB and GS have received reesearch grants and honoraries as speaker and consultant from Novartis Pharma; GR has received honoraries as speaker from Novartis Pharma; FP and GM have received research grants from Novartis Pharma; FR was an employee of Novartis Pharma; CA is an employee of Novartis Pharma.

©Ferrata Storti Foundation

References

1. Weller PF, Bubley GJ. The idiopathic hypereosinophilic syndrome. Blood 1994;83:2759-79.
2. Gotlib J, Cools J, Malone JM, Schrier SL, Gilliland DG, Coutre SE. The FIP1L1-PDGFRα fusion tyrosine kinase in hypereosinophilic syndrome and chronic eosinophilic leukemia: implications for diagnosis, classification, and management. Blood 2004;103:2879-91.
3. Cross NC, Reiter A. Tyrosine kinase fusion genes in chronic myeloproliferative diseases. Leukemia 2002; 16:1207-12.
4. Apperley JF, Gardembas M, Melo JV, Russell-Jones R, Bain BJ, Baxter EJ, et al. Response to imatinib mesylate in patients with chronic myeloproliferative diseases with rearrangements of the platelet-derived growth factor receptor β. N Engl J Med 2002; 347: 481-7.
5. Baxter EJ, Kulkarni S, Vizmanos JL, Jaju R, Martinelli G, Testoni N, et al. Novel translocations that disrupt the platelet-derived growth factor receptor beta (PDGFRB) gene in BCR-ABL-negative chronic myeloproliferative disorders. Br J Haematol 2003; 120: 251-6.
6. Wilkinson K, Velloso ER, Lopes LF, Lee C, Aster JC, Shipp MA, et al. Response to imatinib mesylate in patients with chronic myeloproliferative diseases with rearrangements of the platelet-derived growth factor receptor β. N Engl J Med 2002; 347: 481-7.
7. Pardanani A, Tefferi A. Imatinib targets other than bcr/abl and their clinical relevance in myeloid disorders. Blood 2004;104:1931-9.
8. Grand F, Burgstaller S, Kuhr T, Baxter E, Webersinke G, Thaler J, et al. p53-Binding protein 1 is fused to the platelet-derived growth factor receptor β in a patient with a t(5;15) (q33; q22) and an imatinib-responsive eosinophilic myeloproliferative disorder. Cancer Res 2004; 64:7216-9.
9. Buchdunger E, Zimmermann J, Mett H, Meyer T, Muller M, Druker BJ, et al. Inhibition of the Abl protein-tyrosine kinase in vitro and in vivo by a 2-phenylaminopyrimidine derivative. Cancer Res 1996;56:100-4.
10. Carroll M, Ohno-Jones S, Tamura S, Buchdunger E, Zimmermann J, Lydon NB, et al. CGP 57148, a tyrosine kinase inhibitor, inhibits the growth of cells expressing BCR-ABL, TEL-ABL, and TEL-PDGFR fusion proteins. Blood 1997;90:4947-52.
11. Gleich GJ, Leiferman KM, Pardanani A, Tefferi A, Butterfield JH. Treatment of hypereosinophilic syndrome with imatinib mesilate. Lancet 2002; 359:1577-8.
12. Ault P, Cortes J, Koller C, Kaled ES, Kantarjian H. Response of idiopathic hypereosinophilic syndrome to treatment with imatinib mesylate. Leuk Res 2002;26:881-4.
13. Pardanani A, Reeder T, Porrata LF, Li CY, Tazelaar HD, Baxter EJ, et al. Imatinib therapy for hypereosinophilic syndrome and other eosinophilic disorders. Blood 2003; 101: 3391-7.
14. Cortes J, Ault P, Koller C, Thomas D, Ferrajoli A, Wierda W, et al. Efficacy of imatinib mesylate in the treatment of idiopathic hypereosinophilic syndrome. Blood 2003;101:4714-6.
15. Salem Z, Zalloua PA, Chehal A, Bitar N, Abboud M, Kadri A, et al. Effective treatment of hypereosinophilic syndrome with imatinib mesylate. Hematol J 2003;4:410-2.
16. Baxter EJ, Hochhaus A, Bolufer P, Reiter A, Fernandez JM, Senent L, et al. The t(4;22)(q12;q11) in atypical chronic myeloid leukaemia fuses BCR to PDGFRA. Hum Mol Genet 2002;11:1391-7.
17. Cools J, DeAngelo DJ, Gotlib J, Stover EH, Legare RD, Cortes J, et al. A tyrosine kinase created by fusion of the PDGFRA and FIP1L1 genes as a therapeutic target of imatinib in idiopathic hypereosinophilic syndrome. N Engl J Med 2003;348:1201-14.
18. Pardanani A, Ketterling RP, Brockman SR, Flynn HC, Paternoster SF, Shearer BM, et al. CHIC2 deletion, a surrogate for FIP1L1-PDGFRA fusion, occurs in systemic mastocytosis associated with eosinophilia and predicts response to imatinib mesylate therapy. Blood 2003; 102: 3093-6.
19. Bain B. The idiopathic hypereosinophilic syndrome and eosinophilic leukemias. Haematologica 2004;89:133-7.
20. Klion AD, Noel P, Akin C, Law MA, Gilliland DG, Cools J, et al. Elevated serum tryptase levels identify a subset of patients with a myeloproliferative variant of idiopathic hypereosinophilic syndrome associated with tissue fibrosis, poor prognosis, and imatinib responsiveness. Blood 2003;101:4660-6.
21. Klion AD, Robyn J, Akin C, Noel P, Brown M, Law M, et al. Molecular remission and reversal of myelofibrosis in response to imatinib mesylate treatment in patients with the

myeloproliferative variant of hypereosinophilic syndrome. Blood 2004; 103:473-8.
22. Vandenberghe P, Wlodarska I, Michaux L, Zachee P, Boogaerts M, Vanstraelen D, et al. Clinical and molecular features of FIP1L1-PDFGRA (+) chronic eosinophilic leukemias. Leukemia 2004;18:734-42.
23. Rose C, Dupire S, Roche-Lestienne C, Grardel N, Bourgeois E, Cambier N, et al. Sustained molecular response with imatinib in a leukemic form of idiopathic hypereosinophilic syndrome in relapse after allograft. Leukemia 2004;18:354-5.
24. Martinelli G, Malagola M, Ottaviani E, Rosti G, Trabacchi E, Baccarani M. Imatinib mesylate can induce complete molecular remission in FIP1L1-PDGFR-α positive idiopathic hypereosinophilic syndrome. Haematologica 2004;89:236-7.
25. Pardanani A, Brockman SR, Paternoster SF, Flynn HC, Ketterling RP, Lasho TL, et al. FIP1L1-PDGFRA fusion: prevalence and clinicopathologic correlates in 89 consecutive patients with moderate to severe eosinophilia. Blood 2004;104:3038-45.
26. Musto P, Falcone A, Sanpaolo G, Bodenizza C, Perla G, Minervini MM, et al. Heterogeneity of response to imatinib-mesylate (glivec) in patients with hypereosinophilic syndrome: implications for dosing and pathogenesis. Leuk Lymphoma 2004;45: 1219-22.
27. Roche-Lestienne C, Lepers S, Soenen-Cornu V, Kahn JE, Lai JL, Hachulla E, et al. Molecular characterization of the idiopathic hypereosinophilic syndrome (HES) in 35 French patients with normal conventional cytogenetics. Leukemia 2005; 19:792-8.
28. La Starza R, Specchia G, Cuneo A, Beacci D, Nozzoli C, Luciano L, et al. The hypereosinophilic syndrome: fluorescence in situ hybridization detects the del(4)(q12)-FIP1L1/PDGFRA but not genomic rearrangements of other tyrosine kinases. Haematologica 2005;90:596-601.
29. Muller AM, Martens UM, Hofmann SC, Bruckner-Tuderman L, Mertelsmann R, Lubbert M. Imatinib mesylate as a novel treatment option for hypereosinophilic syndrome: two case reports and a comprehensive review of the literature. Ann Hematol 2006;85:1-16.
30. Pardanani A, Ketterling RP, Li CY, Patnaik MM, Wolanskyj AP, Elliott MA, et al. FIP1L1-PDGFRA in eosinophilic disorders: prevalence in routine clinical practice, long-term experience with imatinib therapy, and a critical review of the literature. Leuk Res 2006;30:965-70.
31. Helbig G, Stella-Holowiecka B, Grosicki S, Bober G, Krawczyk M, Wojnar J, et al. The results of imatinib therapy for patients with primary eosinophilic disorders. Eur J Haematol 2006;76:535-6.
32. O'Brien SG, Guilhot F, Larson RA, Gathmann I, Baccarani M, Cervantes F, et al. Imatinib compared with interferon and low-dose cytarabine for newly diagnosed chronic-phase chronic myeloid leukemia. N Engl J Med 2003;348:994-1004.
33. Rosti G, Martinelli G, Bassi S, Amabile M, Trabacchi E, Giannini B, et al. Molecular response to imatinib in late chronic-phase chronic myeloid leukemia. Blood 2004;103: 2284-90.
34. Mitelman F. An international system for human cytogenetic nomenclature. S.Karger: Basel, 1995.
35. van Dongen JJ, Macintyre EA, Gabert JA, Delabesse E, Rossi V, Saglio G, et al. Standardized RT-PCR analysis of fusion gene transcripts from chromosome aberrations in acute leukemia for detection of minimal residual disease. Report of the BIOMED-1 Concerted Action: investigation of minimal residual disease in acute leukemia. Leukemia. 1999;13:1901-28.
36. Martinelli G, Cilloni D, Rondoni M, et al. Imatinib mesylate for idiopathic hypereosinophilic syndrome (HES). A phase II multicentric Italian clinical trial. Haematologica 2005; 90:Abstract no. 65.
37. Martinelli G, Cilloni D, Rondoni M, et al. Imatinib mesylate can induce molecular complete remission in idiopathic hypereosinophilic syndromem (HES). A phase II multicentric Italian clinical trial. Blood 2005; 106: Abstract no. 375.
38. Rondoni M, Ottaviani E, Piccaluga PP, et al. FIP1L1-PDGFRalpha positive hypereosinophilic syndrome (HES). The response to Imatinib (IM) is durable. A report of 21 patients with a follow-up of 12 to 57 months. Blood 2006;108:7639. Abstract no. 2700.
39. David M, Cross NC, Burgstaller S, Chase A, Curtis C, Dang R, et al. Durable responses to imatinib in patients with PDGFRB fusion gene-positive and BCR-ABL-negative chronic myeloproliferative disorders. 2007;109:61-4.
40. Kerkela R, Grazette L, Yacobi R, Iliescu C, Patten R, Beahm C, et al. Cardiotoxicity of the cancer therapeutic agent imatinib mesylate. Nat Med 2006;12:908-16.
41. Atallah E, Kantarjian H, Cortes J. In reply to 'Cardiotoxicity of the cancer therapeutic agent imatinib mesylate'. Nat Med 2007;13:14 (letter).
42. Rosti G, Martinelli G, Baccarani M. In reply to 'Cardiotoxicity of the cancer therapeutic agent imatinib mesylate'. Nat Med 2007;13:15 (letter).
43. Berman E, Nicolaides M, Maki RG, Fleisher M, Chanel S, Scheu K, et al. Altered bone and mineral metabolism in patients receiving imatinib mesylate. N Engl J Med 2006; 354: 2006-13.
44. Gottschalk S, Anderson N, Hainz C, Eckhardt SG, Serkova NJ. Imatinib (STI571)-mediated changes in glucose metabolism in human leukemia BCR-ABL-positive cells. Clin Cancer Res 2004;10: 6661-8.

©Ferrata Storti Foundation