c-*kit* Mutations in Patients with Childhood-Onset Mastocytosis and Genotype-Phenotype Correlation

Hirokatsu Yanagihori, Noritaka Oyama,
Koichiro Nakamura, and Fumio Kaneko

From the Department of Dermatology, Fukushima Medical
University Graduate School of Medicine, Fukushima, Japan

Mastocytosis represents a clonal proliferation of mast cell hematopoietic progenitors caused by gain-of-function mutations of the *c-kit* gene. The heterogeneity of *c-kit* mutations may have contributed to difficulties in characterizing genotype-phenotype correlation of the disease. Our goal was to analyze a set of reported pathogenic *c-kit* mutations in patients with childhood-onset cutaneous mastocytosis, in comparison with those with adult-onset disease, and to correlate these with clinical presentation. We performed polymerase chain reaction and direct sequencing using genomic DNA samples from 16 nonfamilial Japanese patients with indolent cutaneous mastocytosis (12 with childhood-onset disease and 4 with adult-onset disease) to look for the most common *c-kit* mutations at codons 816, 560, 820, and 839. A substantial number of patients had missense codon 816 mutations (10 of 12 in the childhood-onset group, 83.3%; and 4 of 4 in the adult-onset group, 100%). The most common mutation was Asp816Val (9 of 16, 64.3%) followed by Asp816Phe (5 of 16, 35.7%). Moreover, children with the Asp816Phe mutation developed cutaneous mastocytosis at an earlier age as compared to those with the Asp816Val mutation (mean age of onset, 1.3 months versus 5.9 months, respectively; $P = 0.068$). No other mutation variations were found in our cohort. In summary, we confirmed a high incidence of two distinct *c-kit* mutations, Asp816Val and Asp816Phe, in patients with childhood-onset cutaneous mastocytosis. Our results provide new insights into common *c-kit* mutations, which may contribute to different clinical courses of the disease. (J Mol Diagn 2005, 7:252–257)

Mastocytosis (OMIM 154800) is a diverse group of skin and hematological disorders characterized by hyperproliferative condition of mast cells in various organs, including skin, bone marrow, liver, spleen, and lymph nodes.¹ Onset of disease is usually in infancy or early adulthood. Although most cases of childhood-onset disease resolve spontaneously by puberty,² adult-onset disease can be intractable and may be compounded by systemic symptoms, resulting in mortality. Mastocytosis can be classified into four clinical subgroups: cutaneous disease (IA) or systemic involvement

(IB), disease associated with hematological abnormalities (II), with aggressive mastocytosis (III), and with aggressive carcinomas (IV). IA and IB are the most common phenotypes. The clinical manifestations and prognosis of mastocytosis are difficult to predict and varies considerably between affected individuals.

Molecular-based evidence has shown that this condition results from particular mutations in the proto-oncogene *c-kit* (Figure 1). The gene encodes a transmembrane tyrosine kinase receptor, Kit, that is activated by the binding of specific ligands, such as mast cell growth factor or stem cell factor.³ Kit is expressed on several cell types and is known to activate cell growth and maturation via a phosphorylation-dependent pathway involving tyrosine kinase.

A previous study has shown that there are two major missense changes in the *c-kit* gene at codons 816 and 560, both of which were first identified in the human mast cell line HMC-1.⁴ The former mutation mostly substitutes an aspartate for a valine residue (Asp816Val) in a majority of patients with adult-onset disease, whereas the latter mutation substituting a valine for a glycine residue (Val560Gly) can rarely be detected.⁵ These two missense mutations are also unusual in patients with childhood-onset mastocytosis. Thus, the relationship between the underlining pathogenesis and diverse clinical presentations in mastocytosis may be explained in part by the heterogeneity of *c-kit* mutations.^{5–7} Notably there are no characteristic *c-kit* mutations in patients with childhood-onset mastocytosis.

Recent investigations have disclosed that both Asp816Val and Val560Gly mutations can elicit constitutive activation of the relevant intracellular Kit signaling, resulting in a clonal proliferation of the affected mast cells.⁸ In addition, other pathogenic mutations, Asp816Phe, Asp816Tyr,⁸ Asp816His,⁹ and Asp820Gly,¹⁰ have been shown to induce aberrant autophosphorylation of Kit. However, there is limited information available to understand the functional significance of two *c-kit* mutations: Phe522Cys within the transmembrane portion¹¹ and the dominant inactive mutation Glu839Lys.⁸ Despite peripheral blood and bone marrow integration,

Supported by the Ministry of Education, Science, Sports, and Culture of Japan (grants NO 16790651, KN 14570817, and KN 16591107).

Accepted for publication December 29, 2004.

Address reprint requests to Dr. Hirokatsu Yanagihori, Department of Dermatology, Fukushima Medical University Graduate School of Medicine, 1-Hikarigaoka, Fukushima 960-1295, Japan. E-mail: yanagi@fmu.ac.jp.

Figure 1. Schematic representation of Kit protein and location of *c-kit* gene mutations described previously. The distinct molecular domains comprise three functional structures: NH₂-terminal extracellular, transmembrane and COOH-terminal intracellular domains. The extracellular domain contains five immunoglobulin-like repeats and the intracellular domain encodes two tandem repeats of the enzyme catalytic domains. The mutations at codon 560 within the juxtamembrane region and codon 816/820 within the second catalytic domain cause ligand-independent autophosphorylation of Kit receptor protein, whereas the mutation at codon 839 results in loss of receptor function.⁸ The *c-kit* mutations reported thus far are shown.

these *c-kit* mutations are now considered to be of somatic cell origin.^{8,12} The exact contribution of *c-kit* mutations to the clinical course of mastocytosis remains unclear. In this study, we attempt to characterize the *c-kit* mutation profiles in patients with childhood-onset indolent mastocytosis, and extend genotype-phenotype correlation.

Materials and Methods

Patients

We assessed 16 nonfamilial Japanese patients with cutaneous mastocytosis (Table 1). The cohort included four adults with sporadic disease (one male and three fe-

males; mean age of onset, 29 years), nine children with urticaria pigmentosa (eight males and one female; mean age of onset, 5.7 months), two children with solitary mastocytoma (two males; mean age of onset, 0.5 month), and one boy with diffuse cutaneous mastocytosis (disease onset at 6 months). The former sporadic group comprised three patients with urticaria pigmentosa and one with diffuse mastocytoma. The details are summarized in Table 1. In all patients the diagnosis was confirmed clinically and histologically (hematoxylin and eosin and toluidine blue staining) (Figure 2). After informed consent, biopsy samples were taken from the lesional skin, cut into small pieces, and stored separately for snap-frozen or formalin-fixed paraffin-embedded sections.

Histomorphometric Analysis

Density of mast cells in each skin sample was assessed by toluidine blue.⁵ The number of the mast cells was examined at $\times 400$ magnification (Zeiss, Wetzlar, Germany) and was expressed as the mean cell count per 500 μm^2 from three different grids (Table 1).

DNA Samples

Genomic DNA was extracted from either fresh frozen or formalin-fixed skin sections by a standard proteinase K digestion and subsequent phenol/chloroform extraction method.¹³ The DNA samples were obtained just before the mutation detection procedure.

Mutation Screening

Polymerase chain reaction (PCR) was performed for DNA amplification. Based on the previously published data,¹⁴ the primers used were designed specifically for exons 11, 17, and 18 of the *c-kit* gene (Table 2). The amplification conditions were 94°C for 5 minutes, followed by 40 cycles of 94°C for 30 seconds, annealing temperature (Table 2) for 30 seconds, and 72°C for 30 seconds with a final extension for 10 minutes at 72°C. The reaction mix-

Table 1. Patient Characteristics

Patient	Sex	Age of onset	Clinical diagnosis	Mutation type (codon 816)	Mean dermal mast cell density in lesional skin (number per 500 μm^2)
1	M	1 year	Urticaria pigmentosa	Val	102
2	F	3 months	Urticaria pigmentosa	Val	284
3	M	7 months	Urticaria pigmentosa	Val	204
4	M	3 months	Urticaria pigmentosa	Phe	145
5	M	1 month	Urticaria pigmentosa	Phe	550
6	M	5 months	Urticaria pigmentosa	Val	66
7	M	7 months	Urticaria pigmentosa	Val	148
8	M	6 months	Diffuse cutaneous mastocytosis	Val	544
9	M	Present at birth	Solitary mastocytoma	Phe	672
10	M	1 month	Solitary mastocytoma	Val	71
11	M	1 year	Urticaria pigmentosa	None	350
12	M	1 month	Urticaria pigmentosa	None	406
13	F	59 years	Diffuse cutaneous mastocytosis	Val	77
14	F	28 years	Urticaria pigmentosa	Val	70
15	M	16 years	Urticaria pigmentosa	Phe	74
16	F	13 years	Urticaria pigmentosa	Phe	47

Figure 2. Clinical and histological findings. Our mastocytosis cohort includes three different clinical subgroups: urticaria pigmentosa (case 12, **left column**), diffuse cutaneous mastocytosis (case 8, **middle column**), and solitary mastocytoma (case 9, **right column**). Histology shows marked and dense dermal infiltration of mast cells, although urticaria pigmentosa skin exhibits patchy infiltration in the upper dermis (H&E and toluidine blue staining). Representative clinicopathological features are shown.

ture contains 0.5 ng of individual genomic DNA as a template, 20 pmol of the primers, 2.5 mmol/L $MgCl_2$, 1 mmol/L dNTPs, and 1 U of *Taq* polymerase (TaKaRa Bio Inc., Tokyo, Japan). Aliquots (5 μ l) were purified with a Qiaex II gel extraction kit (Qiagen, Hilden, Germany) and then sequenced directly with an automated DNA sequencer CEQ2000XL (Beckman Coulter, Fullerton, CA) using both the sense and anti-sense primers. To avoid the possibility of introducing PCR errors, both sense and anti-sense readings were repeated at least twice. As a control, archival DNA samples were randomly chosen from unrelated normal patients, and sequenced using the same procedure.

Statistics

Data were analyzed by Student's unpaired *t*-test. A *P* value of <0.05 was considered significant.

Results

Identification of Two Common Missense Mutations in *c-kit*

Assessment by histomorphometry analysis using toluidine blue staining showed that the mast cell density in the lesional skin varied considerably among disease phenotypes (Table 1). However, the minimal cell density (case 16) was sufficient for detecting the mutated *c-kit* allele, thus suggesting adequate amounts of targeting mast cells for the following mutation experiment. For DNA analysis, we designed a new set of PCR primers targeting four independent hot spot *c-kit* mutations reported elsewhere (Table 2). Direct sequencing of amplified DNA from the lesional skin samples identified just two distinct mutations located at the same site in exon 17 (Figure 1); an A>T substitution at nucleotide 2468 that changes an

Table 2. Primer Sequences for DNA Amplification

Primer name	Sequence*	Position	Detection site (amino acid)	Annealing temperature (°C)	Size (bp)
Codon 816/820-F	5'-TGTATTACAGAGACTTGGC-3'	Exon 17	Codons 816 and 820	58	217
Codon 816/820-R	5'-GGATTACATTATGAAAGTCAC-3'	Exon 17			
Codon 560-F	5'-TCTCTCCAGAGTGCTCTAATGA-3'	Exon 11	Codon 560	62	155
Codon 560-R	5'-GGAAGTTGTGTTGGGTCTATGTA-3'	Exon 11			
Codon 839-F	5'-TTCTATTACAGGCTCGACTACCT-3'	Exon 18	Codon 839	60	188
Codon 839-R	5'-GACACCAATGAAACTCAAGATGC-3'	Exon 18			

*Sequence of the primers was designed according to the previous report (GenBank accession number L04143).

Figure 3. Identification of the *c-kit* gene mutation within exon 17. By using genomic DNA from lesional skin, the exon 17 fragment was amplified and sequenced using both sense and anti-sense primers. Representative results from cases 8 and 9 are shown. Sequences of the mutated allele revealed either a substitution from aspartate to valine, Asp816Val (**c** and **d**), or from aspartate to phenylalanine, Asp816Phe (**e** and **f**), but the normal allele did not (**a** and **b**). **Arrows** indicate the nucleotide substitutions.

aspartate residue (GAC) to valine (GTC), Asp816Val, and a doublet transition GA>TT at nucleotides 2467 and 2468 that converts an aspartate residue (GAC) to phenylalanine (TTC), Asp816Phe. Both mutations were heterozygous. None of these patients had other variations of *c-kit* mutations, such as Asp820Gly, Val560Gly, and Glu839Lys. To avoid the possibility of nonpathogenic polymorphisms, it was established that the two codon 816 alterations were absent in 50 ethnically matched control DNA from the Japanese population. As previous reports revealed,^{8,15} we reconfirmed that this was certainly a mutation.

Delineation of Pathogenic c-kit Mutations in Childhood-Onset Mastocytosis

As listed in Table 1, DNA analysis of individual patients disclosed a high incidence of either of two *c-kit* mutations, Asp816Val or Asp816Phe (14 of 16, 87.5%). Of the 12 patients with urticaria pigmentosa, 6 (50%) carried the Asp816Val mutation, 4 (33.3%) the Asp816Phe mutation, and 2 (16.7%) had no hot spot mutations in the *c-kit* gene. One of the two patients with solitary mastocytoma carried the mutation Asp816Val whereas the other carried the mutation Asp816Phe. In contrast, both patients with diffuse cutaneous disease carried Asp816Val. Apart from diffuse disease, there was no significant correlation between particular *c-kit* mutations and phenotypes of mastocytosis (Figure 3).

We further assessed the disease onset on the basis of the two distinct *c-kit* missense mutations, Asp816Val and Asp816Phe. Among 12 patients with childhood-onset dis-

ease, 10 (83.3%) exhibited a codon 816 mutation. Of these patients, seven (70%) had the mutation Asp816Val and three (30%) carried the mutation Asp816Phe. More importantly, the presence of the Asp816Phe mutation correlated with earlier disease onset. Patients with the Asp816Phe mutation developed mastocytosis at a younger age compared to those with the Asp816Val mutation (mean age of onset, 1.3 ± 1.5 months versus 5.9 ± 3.5 months, respectively). However, there was no statistically significant difference between these two groups ($P = 0.068$). In contrast, all four patients with adult-onset disease had codon 816 mutations, two with the mutation Asp816Val and two others with the mutation Asp816Phe (50% in each codon 816 mutation).

Discussion

In this study, we have characterized the common pathogenic *c-kit* mutations in Japanese patients with childhood-onset cutaneous mastocytosis. By using genomic DNA from the lesional skin sites, mutation screening of 16 nonfamilial patients revealed that irrespective of disease phenotype most mastocytosis patients have either Asp816Val or Asp816Phe missense mutations (14 of 16, 87.5%). More importantly, the codon 816 mutations were detectable in most patients with childhood-onset disease (10 of 12, 83.3%). In addition, the mutation Asp816Val, as opposed to Asp816Phe, was predominant among these patients (7 of 10, 70%). In contrast, patients with the Asp816Phe mutation tend to develop cutaneous mastocytosis at earlier age. These findings suggest the pathogenic relevance of the Asp816Phe *c-kit* mutation in the early onset mastocytosis. Likewise, a series of molecular studies may provide a genetic overview of characteristic *c-kit* mutations in cutaneous mastocytosis, as well as the prognostic implications by detection of particular mutations. Nevertheless, we did not identify any *c-kit* mutation variants other than codon 816 in our cohort. Taking into account accumulated data thus far, our results suggest strategies for mutation detection. We propose an initial screening of codon 816 and then the remainder of common *c-kit* mutations in any new patient with childhood-onset cutaneous mastocytosis (particularly in patients 11 and 12 without any hot spot mutations; Table 1). Although our control study includes relatively small number of patients ($n = 50$), they never had polymorphism sequences at codons 816, 820, 560, and 839 in the *c-kit* gene. Furthermore, a series of large cohort studies have demonstrated that these four sites within the *c-kit* gene do not account for polymorphisms.^{8,15-17} Considering these, the possibility for polymorphism at these particular sites can be at least excluded.

Additionally, all patients with diffuse cutaneous mastocytosis carried the Asp816Val mutation (2 of 2, 100%), although there was no statistically significant association between the codon 816 mutations and individual phenotypes. This is in agreement with previous reports showing difficulties in genotype-phenotype correlation, particularly in the heterogeneous observations between human mastocytosis and experimental mice models. A number

of reports have demonstrated germline mutations within exon 11 of *c-kit* in mastocytosis with multiple gastrointestinal stromal tumors.^{16,18} In contrast, Sommer and colleague¹⁹ have failed to reproduce any of clinical phenotypes of cutaneous mastocytosis by introducing the same *c-kit* heterozygous mutation (Val558X) into mice. Further experimental mouse studies may need to be performed to elucidate the relationship between particular *c-kit* mutations and disease phenotypes in mastocytosis.

It should be noted that in our study a relatively higher percentage of patients with childhood-onset mastocytosis carried the Asp816Phe mutation as compared to a previous report.⁸ One possible explanation for this discrepancy is the different procedures used in DNA analysis, ie, direct sequencing, cDNA subcloning and colony sequencing, or restriction enzyme digestion of the amplified PCR products. A more likely reason would be differences in the sample templates or cell sources, such as bulk genomic DNA or *c-kit* cDNA synthesized from lesional skin biopsies, bone marrow aspirates, oral epithelial cells, and peripheral blood cells.^{7,8,17} Furthermore, because *c-kit* mutations in cutaneous mastocytosis are normally a heterozygous state, the detection sensitivity of the particular mutations may depend on the substantial number of local mast cells with the mutated alleles and the amplification rate of the mutated alleles during the initial nonspecific amplification step in PCR. In our study, however, all four adult patients had the codon 816 mutation and specifically, children with the Asp816Phe mutation developed cutaneous mastocytosis at earlier onset than those with the Asp816Val mutation. Therefore, it is conceivable that the codon 816 mutation is not directly related to the particular clinical subtype but perhaps is responsible for earlier onset mastocytosis (particularly less than 1 year) or the prognosis (indolent and transient clinical course).

Although the precise pathogenic relevance of the missense mutation in Kit receptor protein functions are unknown, a number of studies have demonstrated that constitutive autophosphorylation of the Kit signaling pathway by *c-kit* mutations contributes to clonal proliferation of the affected mast cells. This evidence may strengthen the argument for the value of new therapeutic targets relevant to the particular *c-kit* mutations. A potential inhibitor for the Kit signaling, STI-571, prevents the phosphorylation cascade induced by gene mutations within the juxtamembrane domains, such as the Val560Gly mutation, but did not efficiently block the effects of mutations within the enzymatic domains such as the Asp816Val mutation.^{20,21} However, it has been reported that the STI-571 partially inhibits aberrant phosphorylation caused by other mutations, such as Asp816Phe and Asp816Tyr.²⁰ Given these therapeutic advances, accurate mutation detection may be valuable for evaluating the treatment efficacies and management of patients with mastocytosis.

In this study, we have elucidated the molecular basis of cutaneous mastocytosis in 16 nonfamilial Japanese patients. Our data are the first documentation providing the significance of codon 816 *c-kit* mutations in childhood-onset cutaneous mastocytosis, particularly with indolent and transient clinical course. These findings high-

light the fundamental importance of the mutation detection strategy to expand the mutation spectrum in the *c-kit* gene in childhood-onset disease. From the clinical perspective, further investigations regarding a large cohort multicenter study for the identification of pathogenic gene mutations and a longitudinal observation of well-characterized patients are now needed to understand as a yet unidentified disease mechanism in mastocytosis.

Acknowledgment

We thank Dr. Tomoko Inoue (Master of Agriculture, Department of Dermatology, Fukushima Medical University Graduate School of Medicine) for her helpful laboratory work in this study.

References

1. Metcalfe DD: Classification and diagnosis of mastocytosis: current status. *J Invest Dermatol* 1991, 96:2S-4S
2. Azana JM, Torrelo A, Mediero IG, Zambrano A: Urticaria pigmentosa: a review of 67 pediatric cases. *Pediatr Dermatol* 1994, 11:102-106
3. Taylor ML, Metcalfe DD: Kit signal transduction. *Hematol Oncol Clin North Am* 2000, 14:517-535
4. Furitsu T, Tsujimura T, Tono T, Ikeda H, Kitayama H, Koshimizu U, Sugahara H, Butterfield JH, Ashman LK, Kanayama Y, Matsuzawa Y, Kitamura Y, Kanakura Y: Identification of mutations in the coding sequence of the proto-oncogene *c-kit* in a human mast cell leukemia cell line causing ligand-independent activation of *c-kit* product. *J Clin Invest* 1993, 92:1736-1744
5. Buttner C, Henz BM, Welker P, Sepp NT, Grabbe J: Identification of activating *c-kit* mutations in adult-, but not in childhood-onset indolent mastocytosis: a possible explanation for divergent clinical behavior. *J Invest Dermatol* 1998, 111:1227-1231
6. Longley BJ, Metcalfe DD: A proposed classification of mastocytosis incorporating molecular genetics. *Hematol Oncol Clin North Am* 2000, 14:697-701
7. Worobec AS, Semere T, Nagata H, Metcalfe DD: Clinical correlates of the presence of the Asp816Val *c-kit* mutation in the peripheral blood mononuclear cells of patients with mastocytosis. *Cancer* 1998, 83:2120-2129
8. Longley Jr BJ, Metcalfe DD, Tharp M, Wang X, Tyrrell L, Lu SZ, Heitjan D, Ma Y: Activating and dominant inactivating *c-KIT* catalytic domain mutations in distinct clinical forms of human mastocytosis. *Proc Natl Acad Sci USA* 1999, 96:1609-1614
9. Pullarkat VA, Pullarkat ST, Calverley DC, Brynes RK: Mast cell disease associated with acute myeloid leukemia: detection of a new *c-kit* mutation Asp816His. *Am J Hematol* 2000, 65:307-309
10. Pignon JM, Giraudier S, Duquesnoy P, Jouault H, Imbert M, Vainchenker W, Vernant JP, Tulliez M: A new *c-kit* mutation in a case of aggressive mast cell disease. *Br J Haematol* 1997, 96:374-376
11. Akin C, Fumo G, Yavuz AS, Lipsky PE, Neckers L, Metcalfe DD: A novel form of mastocytosis associated with a transmembrane *c-kit* mutation and response to imatinib. *Blood* 2004, 103:3222-3225
12. Vliagoftis H, Worobec AS, Metcalfe DD: The protooncogene *c-kit* and *c-kit* ligand in human disease. *J Allergy Clin Immunol* 1997, 100:435-440
13. Davis LG, Dibner MD, Batte JF: Basic Methods in Molecular Biology. New York, Elsevier, 1986, pp 44-87
14. Kemmer K, Corless CL, Fletcher JA, McGreevey L, Haley A, Griffith D, Cummings OW, Wait C, Town A, Heinrich MC: KIT mutations are common in testicular seminomas. *Am J Pathol* 2004, 164:305-313
15. Longley BJ, Tyrrell L, Lu SZ, Ma YS, Langley K, Ding TG, Duffy T, Jacobs P, Tang LH, Modlin L: Somatic *c-KIT* activating mutation in urticaria pigmentosa and aggressive mastocytosis: establishment of clonality in a human mast cell neoplasm. *Nat Genet* 1996, 12:312-314

16. Nishida T, Hirota S, Taniguchi M, Hashimoto K, Isozaki K, Nakamura H, Kanakura Y, Tanaka T, Takabayashi A, Matsuda H, Kitamura Y: Familial gastrointestinal stromal tumours with germline mutation of the KIT gene. *Nat Genet* 1998, 19:323–324
17. Nagata H, Okada T, Worobec AS, Semere T, Metcalfe DD: c-kit mutation in a population of patients with mastocytosis. *Int Arch Allergy Immunol* 1997, 113:184–186
18. Beghini A, Tibiletti MG, Roversi G, Chiaravalli AM, Serio G, Capella C, Larizza L: Germline mutation in the juxtamembrane domain of the kit gene in a family with gastrointestinal stromal tumors and urticaria pigmentosa. *Cancer* 2001, 92:657–662
19. Sommer G, Agosti V, Ehlers I, Rossi F, Corbacioglu S, Farkas J, Moore M, Manova K, Antonescu CR, Besmer P: Gastrointestinal stromal tumors in a mouse model by targeted mutation of the Kit receptor tyrosine kinase. *Proc Natl Acad Sci USA* 2003, 100:6706–6711
20. Ma Y, Zeng S, Metcalfe DD, Akin C, Dimitrijevic S, Butterfield JH, McMahon G, Longley BJ: The c-KIT mutation causing human mastocytosis is resistant to STI571 and other KIT kinase inhibitors; kinases with enzymatic site mutations show different inhibitor sensitivity profiles than wild-type kinases and those with regulatory-type mutations. *Blood* 2002, 99:1741–1744
21. Zermati Y, De Sepulveda P, Feger F, Letard S, Kersual J, Casteran N, Gorochoff G, Dy M, Ribadeau Dumas A, Dorgham K, Parizot C, Bieche Y, Vidaud M, Lortholary O, Arock M, Hermine O, Dubreuil P: Effect of tyrosine kinase inhibitor STI571 on the kinase activity of wild-type and various mutated c-kit receptors found in mast cell neoplasms. *Oncogene* 2003, 22:660–664