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RESEARCH ARTICLE

A Study on BCR-ABL Kinase Domain Mutations in Chronic Myeloid Leukemia from Western India

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Abstract

Background: BCR-ABL kinase domain(KD) mutations accounts for 60-80% of Imatinib resistance in chronic myeloid leukemia (CML) – chronic phase (CP). Patients with CML who are receiving imatinib treatment, a mutation analysis is required to find out the resistance of imatinib as per European Leukemia Net (ELN) criteria. The present study was carried out to assess for different types of mutations responsible for resistance of imatinib treatment from Western India. **Methods:** In a retrospective study, the patients who were tested for imatinib resistance were analysed for IRMA testing using direct sequencing of BCR-ABL transcript by Sanger method. **Results:** A total of 215 patients were tested for Imatinib resistance analysis (IRMA), of which 45 (20.93%) had detectable mutations. The highest frequency of mutation recorded at T315I amino acids site, followed by M244V and G250E sites. **Conclusion**: The patients who were tested for IRMA showed 20.93 % positive mutations with reference to its resistance are discussed.

Keywords: Imatinib- CML- Sanger's sequencing

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Introduction

Chronic myeloid leukemia (CML) is characterized by positive Philadelphia (Ph+) chromosome. There is reciprocal translocation of t (9;22) (q34;q11) [1] which results in the head to tail fusion of breakpoint cluster region (BCR) gene on chromosome 22q11 with ABL1 (Abelson murine leukemia virus) gene located on chromosome 9q34 creating BCR-ABL1 oncogene. CML has three clinical phases namely chronic, accelerated and blast crises.

The standard treatment of CML-CP is Imatinib mesylate first generation tyrosine kinase inhibitor. Imatinib (400 mg) used as a first-line drug to treat CML which inhibits the abnormal bcr-abl tyrosine kinase inhibitor created by Philadelphia (Ph+) positive chromosome translocation abnormality [2, 3].

It is well established that imatinib still remains gold standard for treatment of CML especially in low income group countries. Although Imatinib is an important treatment of CML but 50-90% of imatinib resistance observed for bcr-abl kinase domain (KD).

Mutation [4]. Therefore, the present study was aimed to assess types of mutations responsible for resistance to imatinib in CML patients of Western India.

Materials and Methods

The samples of patients, who were imatinib resistant were admitted and analysed at S.N.Gene Laboratory and Research Centre, Surat, India. A total of 215 samples received for IRMA analysis were collected between October, 2020 and July, 2021 (ten months) from different parts of Western India. The inclusion criteria were suspected Philadelphia positive patients and exclusion criteria were Philadelphia negative patients. The informed consent was taken from each patients. The IRMA analysis was carried out with Sanger's sequencing method. The results were analysed using BLAST software from NCBI.

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Results

A total of 215 patients were screened, of which 148 were males and 67 were females.

The age varies from 4 to 74 years. Among 215 patients, 170 did not have any type of mutations (Table 1).

The 45 patients who were positive for mutations showed a higher frequency of mutation at T315I, M244V and G250E. In addition, Table 2 shows a list of mutations found in the present study which were resistance to Imatinib, Dasatinib, Nilotinib, Bosutinib and Ponatinib based on the integration published studies between 2001 and 2018 [5].

Table 1. Frequency Analysis of KD Mutations

Number of patients screened	215
Mutation identified in (%)	20.93
Point mutation seen	45
Point mutation seen	Number of patients
T315I	14
M244V	07
G250E	04
F317C	03
F359V	02
D276G	02
H396R	02
F359C	02
F359I	01
S417F	01
M237V	01
M255V	01
E255K	01
Y253H	01
E450K	01
E453V	01
E462K	01

Discussion

Imatinib has shown a success in the treatment of CML in the last few years, there have been reports about Imatinib resistance in overall outcome of disease. The aim of the present study was to investigate the presence of ABL-KD domain mutation in Philadelphia positive (Ph+) cases of CML patients in population of Western India. Of 215 CML Philadelphia positive patients, 45 (20.93%) showed mutations detected by IRMA. Our study is partly in agreement with reported studies in India. Mallekavu et al.[6] have reported a total cases 120, of which, 36 (30%) had detectable mutations. In another study conducted by Rajappa et al. [7] has reported that out of 90 patients, 29 (32.2%) had detectable mutations. On the contrary, a considerably higher percentage of mutations (43%) was reported in the GIMEMA study from Italy [8].

In the present study, we observed a higher frequency of mutations at amino acids T315I followed by M244V and G250E which was in contrast to the study reported by Mallekavu et al. [6] except for a high frequency of mutation at amino acid T315I. We have also compared BCR-ABL KD mutations resistance to Imatinib, Dasatinib, Nilotinib, Bosutinib and Ponatinib based on the integration of published studies between 2001 and 2018 where T315I mutation was resistance to all three generation drugs. The lower percentage of mutations rate (20.93%) was found in the present study could attribute to

Table 2. List of BCR-ABL1KD mutations resistance (+) to Imatinib, Dasatinib, Nilotinib, Bosutinib and Ponatinib based on Integration of Published Study (2001-2018) [5].

Mutation	Imatinib	Dasatinib	Nilotinib	Bosutinib	Ponatinib
T315I	+	+	+	+	+
M244V	+	---	---	---	---
F359I	+	---	+	---	---
F359C	+	---	+	---	---
M237V	+	---	---	---	---
F317C	+	+	---	---	---
G250E	+	---	---	---	---
F359V	+	---	+	---	---
S417F	+	---	---	---	---
D276G	+	---	---	---	---
E255K	+	---	+	+	---
Y253H	+	---	+	---	---
E450K	+	---	---	---	---
E255V	+	---	+	+	---
H396R	+	---	---	---	---
E453V	+	---	---	---	---
E462K	+	---	---	---	---

samples received was primarily of CP-CML.

In conclusion, the patients who were tested for IRMA showed 20.93% positive mutations. The highest frequency was noted in T315I mutation in imatinib treated patients. This will give an indication to clinicians to reconsider therapeutic strategy or go for bone-marrow transplantation.

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Conflict of interest

Authors declare no conflict of interest.

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