doi.org/10.36721/PJPS.2020.33.2.SUP.861-870.1

Long term outcome of chronic myeloid leukemia patients treated with imatinib: Report from a developing country

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Abstract: The outcome of chronic myeloid leukemia has been greatly improved by the use of Imatinib (IM), a selective BCR/ABL kinase inhibitor. The aim of present study was to report long term follow-up & outcome of IM-treated CML patients along with their clinicopathological features, risk group stratification, adverse events and to compare it with CML patients reported from western countries. The mean follow-up of 123 CML patients was 5.5 years in present study, who were treated with frontline IM 400mg daily in a tertiary care hospital in Pakistan. Risk stratification scores, response to treatment (ELN guidelines) and survival outcomes estimated by Kaplan-Meier analysis. Mean age: 35 years (9–67 years) and M: F: 1.5:1, mean follow up time: 5.5 years (1-15 years). Overall survival (OS): at 5.5, 8, 10 and 12 years were 93%, 88%, 81% and 73%, respectively. Progressions free survival (PFS) was 95%, 83%, 83% and 78% at 5.5, 8, 10 and 12 years, respectively. OS estimate by Sokal score was significant (*P-value*: 0.0019). Additional chromosomal aberrations: 1.6%. Eighteen (14.6%) patients progressed to AP/BC. Adverse events were moderate and tolerable. We present findings from a long term follow up of CML patients treated with IM in a developing country. CML mean age at onset was considerably lower than the western populations. Furthermore, 5.5 years OS are comparable to western CML population. IM in our patients as frontline choice proved to be very effective. IM was found to be well tolerated, safe with manageable moderate side effects.

Keywords: Chronic myeloid leukemia, CML, outcome, risk score, survival.

INTRODUCTION

Chronic myeloid leukemia (CML) is a myeloproliferative neoplastic disorder and the hallmark of the disease pathogenesis is characterized by the presence of Philadelphia (Ph) chromosome (Rumpold and Webersinke, 2011). Globally, CML comprises 15-20% of all adult leukemias with an incidence of 1-2/100,000 cases is 1 to 2 cases (Cortes and Kantarjian, 2012). In 2017, it was estimated that 62,130 new cases of leukemia would be diagnosed in United States (US) of which an estimated 9000 would be CML cases, and around 1000 patients would die of CML (American Cancer Society, 2017).

Imatinib mesylate (IM) was introduced in 2001 and the annual mortality after the use if IM has seen a significant decline from 10-20% cases to 1-2% cases annually (Reavie *et al.,* 2013). Previously, the CML cases survival was significantly lower (3-8 years) after onset of the disease (Baccarani *et al.,* 2015). At the time of onset of CML, the median age has been reported to be 55-65 years (Gugliotta *et al.,* 2011; Beinortas *et al.,* 2016) whereas, it was found to present at a younger age in Pakistani patients in earlier studies (Usmani *et al.,* 2009).

The CML treatment and its outcome have improved profoundly by the use of IM, as shown by its promising activity against the disease in phase 2 studies (Cohen *et al.,* 2002). The 10 years survival outcome of CML has improved from 20% to up to 90% (Huang *et al.,* 2012; Deininger *et al.,* 2009). According to the findings of phase 3 study (International Randomized Study of Interferon and STI571: IRIS), the use of 400mg daily in newly onset cases of CML has shown to be more active with negligible side effects as compared to the comparator arm group (interferon-α + cytarabine). Approximately, 76.2% patients in IM arm achieved complete cytogenetic responses (CCyR) at 18 months as compared to 14.5% in the interferon-α + cytarabine arm (P-value: <0.001) while the progression rate was 96.7% in IM arm and 91.5% in interferon-α + cytarabine, (P-value: <0.001) (O'Brien *et al.,* 2003). IRIS trial long term follow up has reported estimated 10 years OS rate of 83.3% in first line IM

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treated CML patients. Overall deaths reported in this study were 86 accounting for 15.6% (n=553). Progression to advanced phases of CML i.e. accelerated and blast phases was seen in 38 patients out of 553 (6.9%). The EFS at 10 years was 79.6% (Hochhaus *et al.,* 2017). The survival rate among CML patients is now determined by coexisting disease conditions rather than by CML (Saussele *et al.,* 2015). Many studies have been conducted to assess and develop the predictive and prognostic models for risk stratification of newly diagnosed CML-CP. Very limited baseline data are available from resource limited countries to inform which risk score can best predict response to treatment and survival outcomes in the IM era (Yamamoto *et al.,* 2014). IM has improved the outcome of CML patients in all phases of the disease. Nevertheless, most of the data in this regard have been reported from the developed countries.

Therefore, the objectives of our study were to study the clinicopathological features of CML patients treated with frontline IM, to report long term outcomes like overall survival (OS) and progression free survival (PFS), to stratify and evaluate the predictive value of 3 risk scores (Sokal, Euro and EUTOS), and to assess the adverse events related to IM therapy.

MATERIALS AND METHODS

Study Plan

It is a single center, observational retrospective study. The blood samples of 123 CML patients were collected between January 2016 to May 2018. The patients' demographics, clinical data and the survival outcomes were analyzed from the date of their diagnosis.

All CML patients were treated in a tertiary care center, Hayatabad Medical Complex (HMC), Peshawar, Khyber Pakhtunkhawa (KP), Pakistan. All patients received IM as first line therapy since 2001 onwards. Prior to data collection, written informed consent was obtained from all patients and the study was conducted according to the "*Declaration of Helsinki*" statements.

Patients with CML were eligible to enter the study if they had a confirmed assay of Philadelphia chromosome positivity (Ph+) from the marrow aspiration, or a positive PCR result for BCR-ABL fusion gene from the blood or BMA. All phases of CML, chronic, accelerated and blast were eligible for enrollment.

The CML patients record from the disease initiation date till last follow up were extracted from the medical charts and recorded in the excel sheet. The parameters recorded included patients' demographics, hematological laboratory investigations, cytogenetic testing and response, FISH assay and response, BCR-ABL quantitative results, medicines prescribed; follow up notes by the clinicians, adverse events (AEs), if progressed to advanced phase (AP/BC) and respective dates, any event related to abnormal hematological, cytogenetic and molecular response, and loss of follow up or death. Patients were evaluated at an interval of 4-8 weeks with complete blood counts, liver and kidney function. Response to therapy was assessed at regular time intervals according to 2013 ELN guidelines (Baccarani *et al.,* 2013).

CML-CP was defined as the presence of less than 5% blast cells, 15 to 19% of basophils, less than 30% blast and promyelocytes cells in the peripheral blood and no evidence of blast cells in extramedullary sites (Cortes *et al.,* 2006). CML-AP was defined as blasts in blood or marrow 15-29% or equal to or greater than 30% blasts + promyelocytes (with blasts count less than 30%) in blood or bone marrow. Beside this, blood basophils equal to or greater than 20% with persistent low platelet counts (<100 $\times$ 10^9/L) and chromosomal abnormalities in Ph+ cells. CML-BP phase was defined by the presence of equal to or greater than 30% blasts in blood or bone marrow with the presence of blast cells in extramedullary sites beside presence in spleen.

Treatment

CML-CP patients received 400mg of imatinib orally, once daily. IM dose was increased to 600-800mg daily was used in IM non-responders or in patients with suboptimal cytogenetic response (minor or minimal) after 6 months. De-novo AP or BC patients were treated with 600-800mg IM daily. Nilotinib was used in CP patients who developed resistance or intolerance to IM and as first line in AP and BC at a an initial dosage of 400mg twice a day and dose was increased up to 600mg to 800mg twice a day, in case of poor response.

Treatment response was determined by examining the patient physically and blood counts were assayed every 4-8 weeks. Aspirates from bone marrow for cytogenetics and differential morphology or FISH tested at diagnosis, 6 month and 12 months, and yearly thereafter. The risk scores were calculated as described in literature (Sokal *et al.,* 1984; Hasford *et al.,* 1998; Hasford *et al.,* 2011). These scores were retrospectively calculated from clinical and laboratory parameters prior to start of IM therapy to categorize patients into different risk groups. CML disease monitoring and response were defined as per ELN guidelines (Baccarani *et al.,* 2006; Baccarani *et al.,* 2013).

Response Criteria

This study objective was to record the type and durability of responses. Cytogenetic testing results of Ph+= 0% or < 1% BCR–ABL nuclei by FISH (out of ≥200 cells) was considered to be equal to complete cytogenetic response (CCyR), the response was considered as partial (PCyR) if the Ph+ chromosomes were 1-35%, minor cytogenetic

response (mCyR) was defined as Ph+= 36-65%, Ph+= 66–95% was considered as minimal cytogenetic response (miCyR) and no cytogenetic response (nCyR) as Ph+= > 95% (Baccarani *et al.,* 2013).

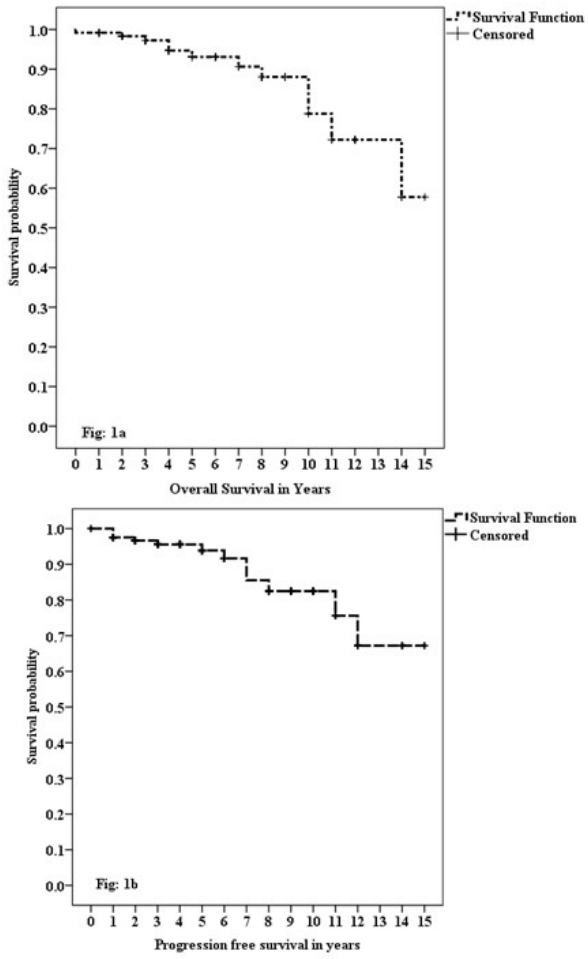


Fig. 1: Kaplan–Meier estimates of fig. 1a: Overall survival, fig.1b progression free survival of 123 patients.

Fluorescent in situ hybridization (FISH) assay from peripheral blood was used to detect BCR-ABL for diagnosis in cases if result of cytogenetics was negative or no metaphase was obtainable. The ELN response to treatment at 3 months as optimal: when Ph+ chromosomes were ≤35%, warning: when Ph+ chromosomes were 36-95% and failure: if the percentage of Ph+ chromosomes were >95% and at 6 months as optimal: Ph+ 0%, warning: Ph+ 1-35% and failure: Ph+ >35%. We were not able to calculate the ELN treatment response at 12 months due to ELN criteria demanding molecular testing for interpretation (Baccarani *et al*., 2013). Molecular responses definition adopted from the updated ELN recommendations. The bcr-abl /abl ratio cutoff of ≤0.1% was defined as major molecular response (MMR), MR4 was defined as a ratio ≤0.01% and MR4.5 was defined as ratio of ≤ 0.0032% (Baccarani *et al.,* 2013).

Secondary endpoints in this study included complete hematological response (CHR), overall survival (OS), and progression free survival (PFS). CHR was defined as white blood cell counts of less than 10,000, with normal distribution of cell types, absence of blast cells, promyelocytes, or myelocytes; normal counts of platelet (150 $X10^9$/L- 450 $X10^9$/L), no evidence of extramedullary disease and disappearance of disease related signs and symptoms.

Survival definitions

(i) OS was determined from the start of initiation of therapy with IM until last follow up date or patient expired date. (ii). PFS was inferred from the IM start date till the documented progression of the CML to AP or BC, or to the date of death; whichever was earlier. Any patient who survived as per last day of study was censored at the last follow-up date. The survival status of lost to follow up patients was confirmed by calling on the registered contact numbers. The survival analysis was determined as per Kaplan-Meier method (Kaplan and Meier, 1958). Hematological and other adverse events were grouped according to the standard terminologies, version 4.03. IM dosage was reduced in those who showed intolerance due to adverse events related to therapy. The treatment with IM was stopped if the grade 3 or 4 toxicity observed. IM therapy was started again at a reduced dose when the toxicity subsided.

STATISTICAL ANALYSIS

We present absolute number and percentages for categorical variables and mean and median with appropriate measure of variation for continuous variables based on the normality test. Comparisons between two groups for categorical data were done using Chi-Square test or Fisher's exact as appropriate. For continuous data we used two sample independent t-test or Mann Whitney U test depending on the normality assumption, where for three or more groups we used analysis of variance (ANOVA) or Kruskal-Wallis test. For time to event outcomes we used Kaplan Meier survival curves and the comparison among groups were done by log rank test. For data analysis we used [SAS/STAT] software version 9.4 (SAS Institute Inc., Cary, NC, USA.) and R foundation was utilized for statistical computing (Vienna, Austria). The Sokal (, Eutos and Euro risk scores were calculated as describes in literature (Sokal *et al*., 1984; Hasford *et al*., 1998; Hasford *et al*., 2011).

RESULTS

One hundred and twenty three patients' medical records were evaluated with initial diagnosis CML-CP treated with frontline IM. Overall characteristics of CML-CP patients' demographics, clinical and laboratory results at baseline are given in table 1. There were more males than

female with a ratio of male to female of 1.5:1. The mean age at diagnosis of CML was 35 (range: 9–67 years). A vast majority (84.6%) of CML patients was in CML-CP phase at time of medical record reviewing, followed by blast phase (10.6%) and accelerated phase accounted for 4.9% of the CML cases. The proportion of female CML patients who conceived during treatment were 8.2% (n=4). Mean follow up time in this study was 66 months (range: 12-180).

During the study period, 6 and 12 patients progressed to AP and BC phase, respectively. The demographics and clinical variables at baseline presentation of CML-CP patients are summarized in table 1. One patient presented with de-novo BC phase. There were 10 confirmed deaths in this study, 9 were related to BC-CML phase and 1 in CP-CML phase (not related to CML). Three patients in CP phase were lost to follow up during the study.

The second generation tyrosine kinase inhibitor (TKI) nilotinib was used in 16.3% of patients who were either IM intolerant or non-responders to IM 400mg daily or escalated dosage. IM dose escalation was made in 19 (15.5%) patients before switching to nilotinib.

Additional chromosomal aberrations (ACAs) were seen in 1.62% of CML patients at baseline. One patient cytogenetically presented 2 Ph+ chromosome, positive for trisomy (+8) and an ISO chromosome, while the other patient's cytogenetics revealed del(6)(q23.1q27).

Risk Scores of CML-CP

The Sokal risk in CML-CP patients was low, intermediate and high in 29 (23.6%), 52 (42.3%), and 40(32.5%), respectively. The Hasford risk score was low, intermediate and high in 40 (32.5%), 51 (41.5%), and 30(24.4%) of CML-CP patients, respectively. The Eutos risk score was low in 100 (81.3%) and high in 23 (18.7%) patients (table 2).

Therapy results of CML-CP

The complete hematologic response (CHR) at 3 months, all treatments, was 93.8% (table 2). Dose reduction was carried out in 4 (3.3%) patients because of low platelet count (1 patient), low hemoglobin and low WBC (1 patient) and pancytopenia (1 patient). Two patients (1.6%) showed IM intolerance and had to discontinue IM. Nineteen patients (15.4%) were documented as non-responder to IM during the treatment. Four patients (3.3%) were non-responder to IM (First line) and nilotinib (second line); all were in blast phase (data not shown in the table).

ELN response CML-CP

The ELN response at 3 months treatment with IM was optimal and warning in 62 (72.9%) and 23 (27.1%) patients, respectively. At 6 months the ELN response was optimal, warning, and failure in 54 (61.4%), 16 (18.2%) and 18 (20.4%) patients, respectively (table 2).

The OS and PFS as determined by Kaplan–Meier analysis is s depicted in fig. 1a and fig. 1b. The Kaplan–Meier estimates of OS at 5.5, 8, 10 and 12 years was 93%, 88%, 81% and 73%, respectively. The Kaplan–Meier estimates of PFS at 5.5, 8, 10 and 12 years was 95%, 83%, 83% and 78%, respectively. The OS probability based on sokal risk score is illustrated in fig. 2. The OS probability based on sokal risk score was statistically significant (Log Rank-Mantel-Cox significance value: 0.007). The difference in OS based on low and intermediate sokal score was not significant statistically (Log Rank-Mantel-Cox significance value: 0.631). Fig. 2b illustrates the OS probability of sokal score based on 2 groups i.e. low risk + intermediate risk score group and high risk score group. The difference in survival probability was statistically significant based on low+intermediate and high score (Log Rank-Mantel-Cox significance value: 0.02).

Characteristics of CML patients who progressed to AP-CML and BC-CML

Patient characteristics at accelerated and blast crisis phase of CML are presented in table 3. In the present study 6 (4.9%) and 12 (9.8%) patients progressed to AP and BC, respectively. The mean age was 29.5 (range 25-46) and 36 (range 16-57) years for AP and BC phase patients, respectively. All CML patients were treated with IM as first line TKI, however, 33.3% of AP and 46.2% of BC patients received second generation TKI, nilotinib, during the course of treatment. The mean time to progression from CP to AP (n=6) was 69.5 (24-147) months and for BC patients (n=12) mean time from CP to AP progression was 58.3 (12-144) months. Mean time to progression from AP to BC for BC patients was 23.5 (5-44) months and mean time from initial diagnosis of CML to BC was 76 (12-161) months. De-novo BC at initial diagnosis was seen in one patient. TKI with chemotherapy was given to 16.7% and 41.7% of AP and BC patients, respectively. After the initial diagnosis, CHR at 3 months was 83.3% and 53.8% for AP and BC CML patients, respectively, while CCyR at 12 months was 83.3% and 58.3% for AP and BC patients, respectively (table 3).

The OS probability of 13 CML-BC patients in months is shown in fig. 3. The mean follow up period was 14.2 months (range: 2-28). The mean OS of BC-CML patients was 14 months with 9 deaths (69.2%) confirmed deaths.

Safety profile

All IM related AEs were not severe, grade 1-2 only, and were well tolerated. The AEs frequency attributable to IM is summarized in table 3. IM related adverse events were documented in 98 patients (79.7%). The most highly reported AEs reported in present study were bone pain and nausea (20.3% each). Hematological AEs (neutropenia, thrombocytopenia and pancytopenia) were documented in 14.6% cases (table 4).

Table 1: Characteristics of CML-CP patients demographics, clinical and laboratory results at baseline (n=123).

Characteristics	# of Patients, %, range
Age	
Mean	35.5 (9-67)
Gender	
Male	74 (60.2)
Female	49 (39.8)
Ratio: Male: Female	1.5-1
CML Phase	
Chronic	104 (84.6)
Accelerated	6 (4.9)
Blast*	13 (10.6)
Hemoglobin (g/dL) Mean	10.1
<12g/dl	69 (56.1)
>12g/dl	14 (11.4)
WBC count ($\times 10^9$/L) Mean	313.7
<50	20 (16.3)
>/=50	64 (52)
WBC-Median, $\times 10^9$/L , range	290 (10.8-666)
Platelets ($\times 10^9$/L) (n =108) Mean	400.2
<450	75 (61)
>/=450	33 (26.8)
Nilotinib used as 2nd Line	
Yes	20 (16.3)
Hydroxyurea	
Yes	52 (42.3)
Interferon	
Yes	3 (2.4)
Received chemotherapy	
Yes	10 (8.1)
Splenomegaly	
<5cm	4 (3.3)
5-8cm	9 (7.3)
>8cm	70 (56.9)
No splenomegaly	40 (32.5)
Hepatomegaly	
Yes	35 (28.5)
Anemia	
Yes	97 (78.9)
Pregnant	
Yes	4 (8.2)
Survival Status	
Confirmed deaths	10 (8.13)
Alive at last follow up	110(89.43)
Lost to Follow up	3 (2.43)
Mean Follow up time, months	66 (12-180)

n= number of patients, WBC: White blood cells count, *= 1 patient presented as De-Novo BP-CML (Blast phase-chronic myeloid leukemia)

Table 2: Risk stratification scores, hematological, cytogenetic response and fluorescent in situ hybridization assay of CML patients to IM therapy (n=123).

Sokal Score	# of patients (%)
<0.8 (low risk)	29 (23.6)
0.8–1.2 (intermediate risk)	52 (42.3)
>1.2 (high risk)	40 (32.5)
Hasford Euro Score	
</=780 (low-risk)	40 (32.5)
>780 and </=1480 (intermediate)	51 (41.5)
>1480 (high-risk)	30 (24.4)
EUTOS Score	
Low risk (</=87 good prognosis)	100 (81.3)
High risk (>87 poor prognosis)	23 (18.7)
CCyR by Bone Marrow Cytogenetics	
CCyR at 3 months	48 (56.5)
CCyR at 6 months	54 (61.4)
CCyR at 12 months	60 (70.6)
CCyR Response by FISH assay	
CCyR at 3 months	43 (51.2)
CCyR at 6 months	37 (44.1)
CCyR at 12 months	47 (56)
CCyR at 18 months	60 (70.6)
Response according to ELN at 3 months	
Optimal (Ph+<35%)	62 (72.9)
Warning (Ph+ 36-95%)	23 (27.1)
Response according to ELN at 6 months	
Optimal (Ph+ 0%)	54 (61.4)
Warning (Ph+ 1-35%)	16 (18.2)
Failure (Ph+ >35%)	18 (20.4)
CHR at 3 months, all treatments	
Yes	106 (93.8)

CHR: Complete hematological response, CML: Chronic Myeloid Leukemia, CCyR: Complete cytogenetic response, FISH: Fluorescent in situ hybridization, ELN: European Leukemia.Net,

DISCUSSION

Tyrosine kinase inhibitors (TKIs) have a very significant role in CML therapy (Cortes, 2012). The treatment goal is to obtain normal blood counts, absence of detectable Ph+ chromosomes cytogenetically and molecular responses at as per time frame of ELN guidelines recommendations. Such guidelines help to guide therapy, and predict the prognosis and long term survival outcomes in CML patients (Haznedaroglu *et al.,* 2013).

There are few reports regarding long term response and survival outcome of CML patients who receive IM as frontline therapy. This is the first report of CML treated with frontline IM from an ethnically distinct population from the northern part of Pakistan. We report the longer term results of IM therapy and outcome. One of the notable finding of our study is the occurrence of CML onset in younger age as compared to western countries (Usmani *et al.,* 2009). This is similar to earlier reports

Table 3: Patient characteristics at accelerated phase (n=6) and blast crisis (n=13) of CML.

	No. (%), or Mean (Range)	
Characteristics	AP (n=6)	BC (n=13)
Gender (Male/Female)	6 / 0	8 (61.5) / 5 (38.5)
Mean Age at AP or BC, years (range)	29.5 (25-46)	36 (16-57)
Hemoglobin , <12 g/dl	4 (66.7)	9(69.2)
White blood cells, >50, $X10^9$/L	1 (16.7)	7(53.8)
Platelets, >450, $X10^9$/L	6 (100)	6 (46.2)
CML phase at initial diagnosis		
CP	6 (100%)	12 (92.3)
BC	-	1(7.7)
Time *CP to BC* diagnosed, months	-	76 (12-161)
Time Progression from *AP to BC*, months	-	23.5 (5-44)
Time *CP* diagnosed to *AP*, months	69.5 (24-147)	58.3 (12-144)
Mean follow up, months	28 (4-41)	14.2 (2-28)
Baseline, CHR*, 3 months	5 (83.3)	7 (53.8)
Initial Cytogenetic Response		
At 6 months (*partial: complete*)	(50%: 50%)	(41.7%: 25%)
At 12 months (*partial: complete*)	(16.7%: 83.3%)	(25%: 58.3%)
Alive/Dead (*Last Follow-up*)	6 (100) / 0 (0)	4 (30.8) / 9 (69.2)

CP: chronic phase, CP-CML: chronic phase-chronic myeloid leukemia, AP-CML: accelerated phase-chronic myeloid leukemia, BC-CML: blast phase-chronic myeloid leukemia, CML: chronic myeloid leukemia, CHR: complete hematologic response

Table 4: Types of adverse events in patients treated with imatinib (n=123).

Type of Adverse Event	Imatinib Treated	
	n	%
Nausea	25	20.3
Bone pains	25	20.3
No Adverse event reported	25	20.3
Hematological toxicity	18	14.6
Hepatotoxicity, abnormal LFTs*	8	6.5
Infections	4	3.3
Gall bladder stones	4	3.3
Pregnancy	4	3.3
Abdominal pain	2	1.6
Diarrhea	2	1.6
Others**	6	4.9

*LFTs: liver function tests like bilirubin, alanine trasferase test (ALT), **others: included, hormonal imbalances like abnormal thyroid stimulating hormone (TSH), gynecomastia, eye edema, ear, drug reaction, skin related (rashes, hypo/hyperpigmentation and pruritus)

from Pakistan and other countries in the region like India and Bangladesh (Gupta and Prasad, 2007; Mottalib *et al.,* 2014). This variation in mean age between the developed and developing countries could be attributed to many factors including age structure of the population, genetic and biological variation of the disease, and more likelihood of younger patients diagnosed and treated.

The CHR rate at 3 months of treatment was 93.8% which is in concordance with a study from India with 3 months CHR rate of 93.8% (Gupta and Prasad, 2007) and another study also reported comparable results (Nair *et al.,* 2014).

The CCyR rate can be seen in the range of 57-88% in CML patients in the TKI era (Haznedaroglu *et al.,* 2014). Conventional cytogenetic testing has an estimated sensitivity of 1-5% (Tefferi *et al.,* 2005). Complete cytogenetics response was noted in 70.6% of CP-CML patients. Our results were in concordance with reports from China on CML patients receiving IM as frontline TKI (Gambacorti-Passerini *et al.,* 2015). The predictive efficacy of risk scores for CCyR was assessed and we noted that Sokal risk score predicted CCyR (p=0.011), while, Euro and EUTOS scores were not predictive of CCyR (p= 0.126 and 0.368, respectively). CML disease

risk scores have been studied by many others (Hasford *et al.,* 2011; Marin *et al.,* 2011) and we noted that there in reports in terms of prognostic and predictive efficacy of these scores. This variation in predictive and prognostic responses from various regions could be explained by many factors like late presentation, genetic differences, and altered drug pharmacokinetics (Kuntegowdanahalli *et al.,* 2016).

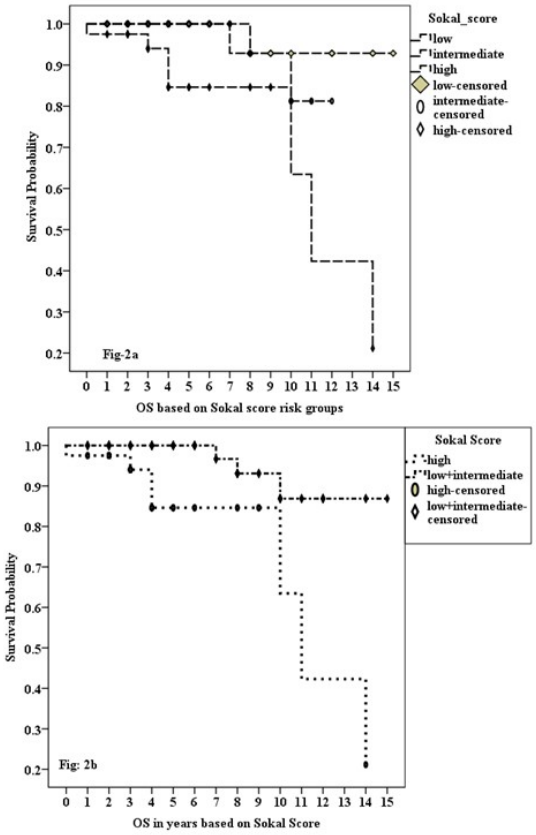
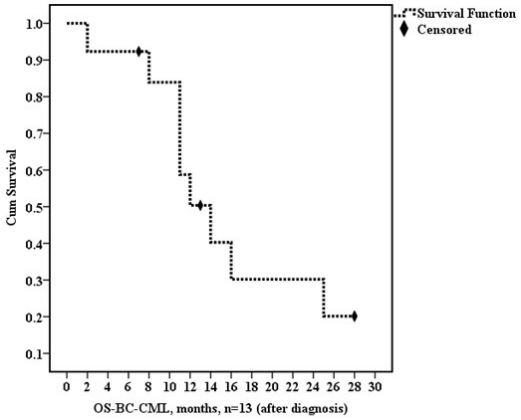


Fig. 2a: Kaplan–Meier estimates of OS probability based on sokal score (lo, intermediate and high risk) and fig. 2b illustrates the OS probability of sokal score (*low+ intermediate score vs. high score*).

The mean follow up time in this study was 5.5 years (range: 1-15, SD ± 3.52). Overall survival in our study cohort was 93% at the end of mean follow up time of 5.5 years. A similar study with a mean follow up time of 5.4 years reported slightly lower OS of 80% (Nair *et al.,* 2014). The 5 years OS from Japan (Ota S *et al.,* 2018) and Lebanon (Massoud *et al.,* 2017) was reported to be 95.1% and 98.3%, respectively. IRIS study reported the estimated 10 years OS rate of first line, IM treated patients as 83.3% (95% CI, 80.1 to 86.6) which is slightly inferior to our results (89%) (Hochhaus *et al.,* 2017). The estimate of survival probability in our present study at 8, 10 and 12 years were 88%, 81% and 73%, respectively.
The PFS probability at 5.5 years was 95% in our study. It was comparable with a study from Tunisia (Ben Lakhal *et al.,* 2018) and USA (Kantarjian *et al.,* 2010), however, inferior PFS has been reported from Lebanon ((Massoud *et al.,* 2017), which can be attributed to a very small number of studied patients (n=46). The estimate of PFS in our study was 83%, 82% and 78% at 8, 10 and 12 years, respectively.



Legend fig. 3: OS: Overall survival, BC-CML: Blast crisis-chronic myeloid leukemia, n= number of patients

Fig. 3: OS in months of BC-CML patients (n=13).

In our study, the factors that were associated with significantly favorable OS outcome included CHR at 3 moths, cytogenetic response, ELN response at 3 and 6 months and sokal score. A stable response on FISH study was significantly associated with longer OS rate (*p= 0.012*).

During the follow up period, 18 (14.8%) patients progressed to AP or BC. In one study progression to CML advanced phases (accelerated or blast crisis) was noted in 38 patients out of 553 (6.9%) (Hochhaus *et al,* 2017). Progression to advanced phase in high percentage of our patients could be attributed to late presentation, patients wasting time with quacks and non-specialists, and poor compliance to therapy.

Additional chromosomal aberrations (ACAs) were seen in 2 patients at the time of diagnosis. ACAs are generally associated with decreased response to TKI therapy and poorer survival. **(**Baccarani *et al.,* 2013). One of our CML patient at baseline had *double Ph+ chromosome*, trisomy (+8) in all cells and an ISO chromosome, i(17)(q10). This patient was having 2-ACAs of Group 1(favorable prognosis group) and 1-ACA of group 2 (poor prognosis group) as described in literature (Wang *et al.,* 2016). This patient had a good response to therapy. The second patient had ACA at baseline showing del (6). The del (6) is classified as minor route ACAs and has been rarely reported previously (Su *et al.,* 1999). This patient also had a favorable response to treatment.

IM was well tolerated in our patients with over all good compliance. Bone marrow related complications like neutropenia, thrombocytopenia, and pancytopenia were documented in 14.6% of IM treated cases. Some studies

have documented hematological AEs related to TKIs as risk factor of long term survival (Sneed *et al.,* 2004, Funke-Vaneuza *et al.,* 2005).

Four CML-CP patients conceived during therapy. All of them were on IM 400mg/day. Three patients received interferon in first trimester and then resumed IM 400mg/day. One of them was non responder to IM dose was increased up to 800mg/day and was shifted to nilotinib 400mg twice a day. This patient achieved CHR and complete cytogenetic response and was doing well at last follow up.

The forth patient was having poor compliance to regular intake of IM and during pregnancy remained on IM 400mg/day due to high TLC. After one year post-delivery, she progressed to AP. IM dosage was escalated up to 800mg but the patient progressed to BC after 8 months, received chemotherapy with an initial good response but relapsed again and died after 13 months. All 4 patients gave birth to normal healthy babies without any complications.

In the patients with CML in AP CHR was similar to the previous reports, however, our complete cytogenetic responses and OS were higher than previously reported (Aziz *et al.,* 2007; Kantarjian *et al.,* 2005) Similarly, in BC patients, the CHR, cytogenetics and survival outcome was superior than reported in earlier studies (Palandri *et al.,* 2008; Aziz *et al.,* 2007).This higher response and survival in AP/BC patients may have been due to younger age at presentation, small number of patients, early presentation, optimal compliance and clonally stable genome from ACAs. Several factors can be attributed to the high response rates in our CML-BC patients, namely; early disease presentation, statistically small number of patients, very young age at presentation and less advanced phase of disease clonally.

Just like any study, our study main limitations were retrospective nature, small sample size and lack of molecular testing due to non-affordability of RT-PCR assay cost for BCR-ABL (~146$).

CONCLUSION

In summary, we present the findings of long term follow up (5.5 years) of CML patients from Pakistan, treated with frontline IM. The mean age of our studied cohort at CML diagnosis was significantly younger than patients from developed countries. Only Sokal score had prognostic prediction for OS (p=0.0019) with overall inferior survival among patients with intermediate and high sokal score as compared to low risk patients. Sokal risk category also predicted CCyR with low risk patients having better responses (p=0.011). In conclusion, IM use as frontline medication for CP-CML was found to be effective, safe and well tolerated with moderate manageable side effects.

ACKNOWLEDGEMENTS

Our special thanks to the staff of Hayatabad Medical Complex mainly Mr. Ikram Khan and Mr. Wahab Khan for their cooperation and hospitality. We further wish to thank Mr. Waseem Sajjad and Mr. Ghani Rehman for their help in drawing the blood samples for molecular studies.

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