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Überörtliche Berufsausübungsgemeinschaft
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Laboratory information 4/2017

Molecular and cytogenetic BCR-ABL1 follow up of TKI-treated chronic myeloid leukemia (CML)

Medical background

The well-known **BCR-ABL1 oncogene transcripts** deriving from a t(9;22)(q34;q11) translocation are the hallmark of CML. Over the last decades, the survival of patients with CML has dramatically improved. The last big step of this story was the introduction of tyrosine kinase inhibitors beginning with Imatinib (Gleevec) in 2001. Since then, **molecular follow up** has become an integral part of the European LeukemiaNet recommendations for the management of chronic myeloid leukemia[1], as well as reflected at the arab leukemia net[2]. Hematologic, cytogenetic and molecular follow up allows for stratifying patients into "optimal response", "warning" and "failure" groups, respectively, with different long term outcome. Patients with optimal response, e.g. at least a major molecular response (MMR) when PCR results are expressed on the "**international scale**", show the best long-term outcome so there is no need for a change of treatment. In case of "failure", patients should receive a different treatment to limit the risk for progression and death. When the follow up points to "warning", more frequent monitoring is recommended to enable timely treatment decisions in case of treatment failure. A pocket card for medical doctors[3] containing all valuable information for TKI treated CML patients is available from the European leukemia net (here partly shown).

ELN European LeukemiaNet — UPDATE 2013

European LeukemiaNet Recommendations for the Management of Chronic Myeloid Leukemia (CML)

Baccarani et al, Blood 2013;122:872-884

Response definitions for any TKI first line, and 2nd line in case of intolerance, all patients (CP, AP, and BC)

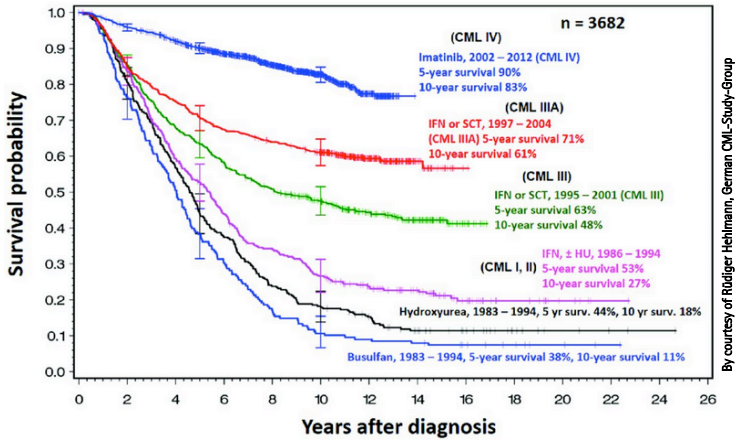
Time	Optimal response	Warning	Failure
Baseline		High risk Major route CCA/Ph+	
3 mos.	BCR-ABL^{IS} ≤10%* Ph+ ≤35% (PCyR)	BCR-ABL^{IS} >10%* Ph+ 36-95%	No CHR* Ph+ >95%
6 mos.	BCR-ABL^{IS} <1%* Ph+ 0% (CCyR)	BCR-ABL^{IS} 1-10%* Ph+ 1-35%	BCR-ABL^{IS} >10%* Ph+ >35%
12 mos.	BCR-ABL^{IS} ≤0.1%* (MMR)	BCR-ABL^{IS} 0.1-1%*	BCR-ABL^{IS} >1%* Ph+ >0%
Then, and at any time	MMR or better	CCA/Ph- (-7, or 7q-)	Loss of CHR Loss of CCyR Loss of MMR, confirmed** Mutations CCA/Ph+

*and/or **in 2 consecutive tests, of which one ≥1% IS: BCR-ABL on International Scale

Timing of Cytogenetic and Molecular Monitoring

At diagnosis	CBA, FISH in case of Ph- (for cryptic or variant translocations), qualitative PCR (transcript type)
During treatment	RQ-PCR every 3 months until MMR has been achieved, then every 3 to 6 months and/or CBA at 3, 6, and 12 months until CCyR has been achieved, then every 12 months. Once CCyR is achieved, FISH on blood cells can be used.
Failure, progression	RQ-PCR, mutational analysis, and CBA. Immunophenotyping in blast phase.
Warning	Molecular and cytogenetic tests more frequently. CBA in case of myelodysplasia or CCA/Ph-

CBA: Chromosome banding analysis of marrow cell metaphases at least 20 metaphases analysed



The timing of additional investigations depends on the course of follow up parameters. An occurrence of additional clonal aberrations CCA (cytogenetics), which are present in 9.3% of egyptian patients[4], or new **point mutations** within the BCR-ABL1 tyrosine kinase domain (sequencing) may guide or enable important treatment decisions, e.g. to change to a second-/third-line TKI in case of secondary resistance or to search for a donor when transplant is an option.

Other definitions

CCA	Clonal chromosome abnormalties
CCA/Ph+	CCA in Ph+ cells which define failure if newly arisen
CHR	Complete hematologic response: Platelet count < 450 x 10^9 /L; WBC count <10 x 10^9 /L; Differential: no immature granulocytes, basophils <5%; no palpable spleen
High risk	Evaluated by Sokal-Score (>1.2), Euro-Score (>1,480) or EUTOS-Score (>87)
Major route CCA/Ph+	Major route CCA/Ph+ are trisomy 8, 2nd Ph+ [+der(22)t(9;22)(q34;q11)], isochromosome 17 [i(17)(q10)], trisomy 19, and ider(22)(q10)t(9;22)(q34;q11)
Mutations	BCR-ABL kinase domain point mutations (not to be confused with ABL1 polymorphisms), Mutational analysis by conventional Sanger sequencing is recommended in case of progression, failure and warning.

Method, turnaround time, reporting

Quantitative RT-PCR, FISH, cytogenetics. Allow three days for RT-PCR. For each medical report, we supply an interpretation along with further follow up recommendations.

Material

Quantitative RT-PCR or FISH: peripheral EDTA whole blood, 10 ml
cytogenetics: lithium-heparinized bone marrow, 2ml

Contact

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Literature

1. Baccarani et al., BLOOD, 8 August 2013, Volume 122, No. 6, 872-884
2. http://www.aln-afme.com/
3. https://www.leukemia-net.org/content/leukemias/cml/recommendations/e8078/infoboxContent10432/PocketCard_UPDATE2013_English.pdf
4. Azzazi et al., Blood 2014 124:5539

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