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Research in the heart of hematology: chronic myeloid leukemia 2017

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One of the great success stories of modern hematology is reaching its next and possibly final phase: the achievement of treatment-free remissions in stable deep molecular responders with chronic myeloid leukemia (CML) which may well be equivalent to cure. Although only the minority of patients achieve treatment-free remissions, the absolute numbers of patients currently in discontinuation studies (Table 1) and in durable treatment-free remissions (40- 60%) are impressive and argue for a change in the treatment strategy for CML. The progress since last year cannot be overlooked.[1] The goal is to define patients in whom treatment can be stopped safely and to establish a strategy for treatment discontinuation.[2]

This is not the first amazing success in some 50 years of basic and clinical research underlying the success story of CML: the detection of oncogenes and of kinase activity in many of them was fortuitous, since it was a byproduct of the search for human leukemia viruses. In realization that most animal leukemias could be induced by viruses, this was a high priority research field in the late 1960s and early 1970s. Large national programs funded with billions of dollars, such as the Special Virus Cancer Program and the National Cancer Act for the "conquest of cancer", had been started in the USA. With modern molecular biology methods, so-called footsteps of viruses were looked for. The detection of reverse transcriptase in human leukemic cells[3] and of virus-related RNA and DNA in human cells and in the human genome[4,5] were at the time interpreted as breakthroughs on the path to detection of human leukemia viruses. Whereas ultimately no such viruses were found associated with common human leukemias, oncogenes proved central in human carcinogenesis. An example is the role in CML of the *ABL* oncogene which, in 1980, was detected in the acutely transforming defective Abelson leukemia virus in which parts of the virus genome had been replaced by cellular sequences.[6] It was shown that most retroviral oncogenes were present as so called protooncogenes in the human genome pointing early to ubiquity and important functions of these genes in

the biology of normal and malignant cells. The discovery that the *ABL* oncogene was located on chromosome 9 at the breakpoint of the t(9;22) translocation,[7] and of fusion transcripts of *ABL* with the *BCR* region on chromosome 22[8] paved the way to the stunning observation that *BCR-ABL* sequences could induce leukemia in mice.[9,10] Since *ABL*, like many other oncogenes, had tyrosine kinase activity, legions of tyrosine kinase inhibitors (TKI) were produced.[11] It was the logical next step to define an inhibitor specific for *BCR-ABL* and suitable for therapeutic use in humans.[12]

The current global strategies are aimed at recognizing patient- and treatment-related factors indicating that treatment discontinuation would be successful and safe. Optimization of current treatments has priority over the development and characterization of new drugs. New and better drugs may become more important again when strategies for TKI discontinuation have been optimized and the specific needs for drug treatment of patients who do not qualify for discontinuation are better known. More than 20 studies on treatment discontinuation have been published and even more were submitted for presentation at the 2016 American Society of Hematology (ASH) annual congress (Table 1). The total number of patients in published and ongoing studies on this subject is well above 3000. The largest of the studies, the EURO-SKI study,[13] reports on 750 TKI (mostly imatinib) pre-treated patients with a follow-up after discontinuation of up to 36 months. Evolving factors that have been identified to predict successful discontinuation and stable treatment-free remissions are duration of TKI treatment (≥5.8 years better) and duration of deep molecular response (each additional year increases the probability of staying in major molecular remission by 16%). The impact of high Sokal risk score, younger age, gender, prior suboptimal response, TKI resistance, line of therapy, depth of remission (molecular response *versus* greater than molecular response) and other factors require confirmation or longer follow-up in larger cohorts. Treatment discontinuation can even be successful at a second attempt: based on a study of 60 patients, Legros *et al.* reported that the chances of success are not much different from those after first attempts.[14]

It came as a surprise that treatment discontinuation may induce adverse effects and that quality of life before and after stopping TKI treatment may not be much different. The condition, which is termed TKI-discontinuation syndrome,[15] with joint and muscle pain resembles polymyalgia rheumatica and occurs in about 30% of patients. In the majority of cases it seems to subside after some time and rarely requires reinstitution of TKI treatment.

An interesting observation is that no type or dose of TKI has thus far been shown to produce a clear survival advantage. An explanation could be that current TKI treatment is so efficient and survival so close to that of the general population that further improvement becomes difficult to prove, particularly in view of the fact that currently more patients die of comorbidities than of CML. Proof for this needs to be obtained by long-term observation of sufficiently large cohorts with survival as an endpoint.

An apparent limitation of progress is the concern that the same quality of CML management is not provided everywhere, not even in Europe or North America. The European LeukemiaNet (ELN) management recommendations for CML try to provide uniform definitions and rec-

Table 1.

Study	TKI	Min. treatment duration (years)	N.	Depth of MR	Min. duration of MR (years)	RFS with at least MMR
Euro-SKI[1]	IM	3	750	MR[4]	1	52% at 2 years
STIM[2]	IM	2	100	Not determined individually	2	38% at 7 years
TWISTER[3]	IM	3	40	MR[4.5]	2	45% at 42 months
A-STIM[4]	IM	3	80	UMRD*	2	64% at 23 months
KIDS[5]	IM	3	90	MR[4.5]	2	58% at 2 years
HOVON[6]	IM	20 months	18	MR[4.5]	2	33% at 3 years
STIM2[7]	IM	2	200	MR[4.5]	2	46% at 2 years
ISAV[8]	IM	2	108	UMRD*	1.5	52% at 22 months
STOP 2G-TKI[9]	DAS / NIL	2	60	MR[4.5]	2	≈ 55% at 4 years
DADI[10]	DAS 2nd line	ND*	63	MR[4]	1	49% at 6 months
NILST[11]	NIL	2	87	MR[4.5]	2	59% at 1 year
TRAD[12]	IM / DAS	3	75	MR[4.5]	2	58% at 6 months
Dasfree[13]	DAS	2	130	MR[4.5]	1	63% at 1 year
ENESTop[14]	IM / NIL	3	126	MR[4.5]	1	58% at 4 years
STAT2[15]	IM / NIL	2	96	MR[4.5]	2	68% at 1 year
ENEST freedom[16]	NIL	3	190	MR[4.5]	1	52% at 4 years
D-STOP[17]	IM / DAS	ND*	54	MR[4]	2	63% at 1 year
RE-STIM[18]	(2nd stop)	35 months	67	mostly UMRD*	31 mos	44% at 22 months
Total: 18			**2334**			**33% – 68% after 0.5 – 7 years**

TKI: tyrosine kinase inhibitor; IM: imatinib; DAS: dasatinib; NIL: nilotinib; *ND: not defined, MR: molecular response, UMRD: undetectable minimal residual disease; RFS: relapse-free survival; MMR: major molecular response. [1]Mahon *et al.*, ASH-Abstract 2016; [2]Mahon *et al.*, Lancet Oncology 2010;11:1029; [3]Ross *et al.*, Blood 2013;122:515; [4]Rousselot *et al.*, JCO 2014;32:424; [5]Lee *et al.*, Haematologica 2016;104:717; [6]Thielen *et al.*, EJC 2013;49:3242; [7]Nicolini *et al.*, ASH-Abstract 2013 # 654; [8]Mori *et al.*, AJH 2015;90:910; [9]Rea *et al.* Blood 2016, epub ahead of print; [10]Imagawa *et al.*, Lancet Haematol. 2015; [11]Kadowaki *et al.*, ASH-Abstract 2016; [12]Kim *et al.*, ASH-Abstract 2016; [13]Shah *et al.* ASH-Abstract 2010; [14]Hughes *et al.*, ASH-Abstract 2016; [15]Takahashi *et al.* ASH-Abstract 2016; [16]Hochhaus *et al.*, ASCO-Abstract 2016; [17]Keenajai *et al.*, ASH-Abstract 2016; [18]Legros *et al.*, ASH-Abstract 2016.

ommendations globally. The population-based registry of the European Treatment and Outcome Study (EUTOS) for CML project, a public-private partnership between the ELN and Novartis, now reports that in 20 European countries most patients are managed according to ELN recommendations.[16] The prospects are therefore excellent that in Europe the new treatment discontinuation strategy in CML will be available to most CML patients even in routine care. The reliable availability of high quality, standardized, molecular monitoring is of the utmost importance.

Another registry study (Simplicity) involving 1,494 TKI-treated CML patients from North America and Europe found lower rates of molecular monitoring in the USA than in Europe. In an analysis of switching therapies within the first 12 months, intolerance of first-line TKI was, at >70%, the most frequent reason for switching treatment.[17]

Suboptimal tolerability of TKI and adverse effects, particularly of second- and third-generation TKI, may be obstacles to achieving the best possible outcome. The ELN has, therefore, appointed an international panel of experts, including experts from North America and Asia, to provide evidence-guided recommendations for the management of TKI-related adverse events.[18] The recommendations will help to provide uniform and high quality management of CML patients globally.

In spite of the excellent long-term tolerability of imatinib, careful monitoring for late toxicities remains important. Some reports indicate that kidney function should be more carefully monitored since a decrease of glomerular filtration rate has been observed after long-term imatinib treatment. Older age and lower estimated glomerular filtration rate at the initiation of imatinib were found to be associated with later development of chronic kidney disease.[19] This confirms an earlier report by Marcolino *et al.*[20] Since nilotinib has been reported beneficial for renal function, a switch to nilotinib may be considered in patients with these characteristics.

Pricing has been a topic of serious concern in CML treatment for some time. The now general availability of generic imatinib alleviates this concern, but the question remains whether the quality of generic imatinib preparations will be equal to that of branded imatinib. At least five contributions to the ASH 2016 conference have addressed this question and agree that generic imatinib is of similar quality. The largest cohort was from the Polish Imatinib Generics Registry: Sacha *et al.* prospectively observed 726 patients treated with various generic imatinib preparations (mostly Nibix and Meaxin) for 1 year and concluded that the clinical efficacy and tolerability of the tested generics are not inferior to those of branded imatinib.[21]

A long-neglected, but central determinant of the natural course of CML is additional chromosomal aberrations as a consequence of *BCR-ABL*-induced genetic instability. After the frequency of such aberrations in blast crisis had been observed[22] and the relevance of clonal evolution for progress of CML recognized, more detailed information was provided recently on which additional chromosomal aberrations may be drivers and which are merely bystanders.[23] Fabarius *et al.* analyzed the prognostic impact of unbalanced additional chromosomal aberrations at

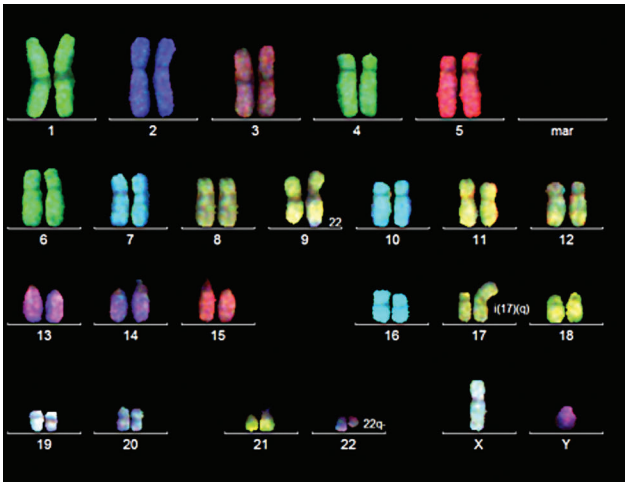

Figure 1. Translocation t(9;22) with iso-chromosome i(17)(q10) which has been identified as a poor prognosticator at diagnosis and in the course of CML. The long arm of chromosome 17 is doubled and replaces the short arm in an inverse direction. Multi-color fluorescence *in situ* hybridization. Courtesy of C. Haferlach and A. Fabarius.

diagnosis and found that only major route aberrations (+8, +Ph, i(17)(q10) and +19) had a negative impact on survival.[24] Wang *et al.* determined the impact of additional chromosomal aberrations arising *de novo* in the course of CML in more than 2,000 patients and categorized the aberrations according to their impact on survival. The most unfavorable were chromosome 17, 7 and 3 aberrations [i(17)(q10), 3q26, -7] (Figure 1). Trisomy 8 was found to be less unfavorable unless it was combined with other additional chromosomal aberrations.[25] Chen *et al.* examined the differential impact of additional chromosomal aberrations on lymphoid and myeloid blast crisis and found that such aberrations confer an inferior prognosis in myeloid, but not in lymphoid blast crisis.

The challenge for the coming years will be how best to prevent additional chromosomal aberrations in the remaining patients who cannot be treated satisfactorily with TKI.

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Research in morphology and flow cytometry is at the heart of hematology

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Through its inherent implication in such major physiological systems as oxygenation, coagulation, protection against infections and tumor proliferation, hematology is a nearly boundless field for research and discovery. Indeed, hematology has been at the heart of such fundamental work as the understanding of iron regulation in red blood cell mediated tissue oxygenation,[1] or the deciphering of the intricate molecular interactions between endothelial cells, platelets and plasmatic proteins in the early stages of hemostasis.[2] Deeper insights into cell biology have also been attained through the use of the easily available blood or bone marrow cells and cell lines derived from hematological malignancies. Moreover, the notion of the importance of the microenvironment of many cells has certainly found fertile ground in the study of the bone marrow or diseased lymph nodes involved in leukemia and myeloproliferative or lymphoproliferative disorders.[3-5] Hematology is thus entertaining strong transdisciplinary interactions with pathology, immunology, biochemistry, cytogenetics, molecular biology and also, more recently, advanced imaging techniques.[6] These various aspects deal with the physiological maintenance of homeostasis, a precise and tightly regulated phenomenon in an extremely active system producing and eliminating trillions of cells every day. They are also at the heart of our increasing understanding of the mechanisms of disease and targeted therapeutic approaches.

In the field of hematology, two sub-disciplines are at the basis of diagnosis for malignant and non-malignant processes, i.e., the morphology of hematopoietic cells and flow cytometry. Both are considered indispensable, but it is sometimes forgotten that both represent true specialties, requiring thorough training and experience. It could be argued that the progresses of automation could alleviate these prerequisites, but the reality seems more subtle. Indeed, blood cell counters performing a complete blood count (CBC) are becoming more and more sophisticated, as previously underscored in the 2016 editorial from our scientific working group on innovation.[7] With different technological approaches, latest generation instruments perform accurate and reproducible quantitative measurements of peripheral blood cell composition, and have good sensitivity and specificity for flagging the presence of abnormal cells.[8] Understanding the flags or messages generated by these intelligent machines, in particular through morphological cell identification on a stained blood smear, still requires the knowledge initially placed in the design of automated interpretation. In addition, as with most expert systems, when the machine is at a loss, only the brain of the biologist/hematologist can come to the rescue, a task made increasingly difficult by the lack of experience in normal or benign situations taken care of by the instrument. Furthermore, cytomorphological analysis of bone marrow aspirates remains a cornerstone in the most recent WHO classification, and requires additional skill and knowledge.

The same is true for flow cytometry, with new instruments managing the whole process of handling samples