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EORTC Leukemia Group achievements

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ABSTRACT

The EORTC Leukemia Group (LG) has a long history of promoting the study of leukemias and related malignancies and reports here on three of their most significant achievements. In acute myelogenous leukemia (AML), the LG and Italian group GIMEMA started their fruitful collaboration in 1986 with the AML-8 trial with 1519 inclusions. In the AML-8A trial, in patients who reached complete remission, without a HLA identical sibling, autograft provided longer disease-free survival than a second course of consolidation, whereas the best outcome was observed in patients with a donor, who had to be allografted. The AML-10 trial set a new standard of treatment for induction/consolidation with replacement of daunorubicin by either idarubicin or mitoxantrone. The AML-12 trial tested the effect of high-dose cytosine-arabinoside during induction (2109 inclusions, data base locked in August 2011 for final analysis). Development of intergroup trials focusing on subgroups of AML bearing specific genetic abnormalities is now mandatory to validate the “targeted approach” of driving molecular events. In high-risk myelodysplastic syndrome (MDS), the phase III trial conducted by the LG in collaboration with the German MDS Study Group showed that the response rate of decitabine versus best supportive care was higher (complete or partial remissions, 19% versus 0%, and hematologic improvement, 15% versus 2%), progression-free survival was significantly prolonged (median 6.6 versus 3 months), cumulative incidence of AML was significantly decreased

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(22% versus 33% at one year), but the impact on OS was less evident (median 10.1 versus 8.5 months; hazard ratio 0.88). Quality of life had improved significantly in patients in the decitabine arm. The assessment of HDAC inhibitors, such as vorinostat, will probably be tested in the next trial. Also in MDS, relevant genetic lesions involved in the pathogenesis of this disease were identified using single nucleotide polymorphisms array-based genomic profiling and genomic sequencing in 102 patients with MDS. Acquired abnormalities of the TET2 gene were identified in 26% of the cases and in the EZH2 gene in 5–10% of the patients. TET2 mutations were detected in 96% of the bone marrow cells, including CD34+ progenitor cells, suggesting that TET2 mutations could be an early event during disease evolution. In normal bone marrow, TET2 expression was elevated in granulocytes, suggesting a role in myelopoiesis.

Conclusion: during the last 25 years the EORTC LG in cooperation with GIMEMA made a considerable contribution to the improvement of treatment results of patients with acute leukemia or MDS.

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1. Introduction

The aim of the EORTC Leukemia Group (LG) is to promote the study of leukemias and related malignancies with regard to their natural history, diagnosis, treatment and sequelae of treatment. Specifically, the LG organizes phase I, II and III trials for patients with myeloid or lymphoid leukemias and myelodysplastic syndromes (MDS).

2. Optimizing treatment in acute myelogenous leukemia

The treatment of acute myelogenous leukemia (AML) relies on two active drugs used in this disease dating back 40 years: daunorubicin, an anthracycline, being a DNA intercalating agent, associated with cytosine arabinoside, an antimetabolite, for induction treatment, followed by the same association using cytosine arabinoside at higher dose (consolidation). According to cytogenetic and molecular biology, intensification could be allograft, autograft, or courses of high doses of cytosine arabinoside. With the induction treatment, approximately 60% to 80% of adults with AML achieve complete remission (CR). Strategies to improve anti-tumor efficacy have included the addition of a third drug (etoposide) and the substitution of daunorubicin by other anthracyclines: idarubicin or mitoxantrone. However, because of the relatively small sample size and/or the relatively short follow-up time, the benefits of these new agents on the long-term outcome of patients with AML have not been definitively established.

The aim of the EORTC LG and GIMEMA AML-10 trial was to compare the efficacy and toxicity of three different anthracyclines in combination with cytarabine and etoposide in adult patients with newly diagnosed AML. This study randomly assigned 2,157 patients (age range 15 to 60 years) to receive intensive

induction–consolidation chemotherapy containing either daunorubicin, or idarubicin, or mitoxantrone. After achieving CR, patients were assigned to undergo either allogenic or autologous stem-cell transplantation (SCT) depending on the availability of a sibling donor. The overall CR rate (69%) was similar in the three groups. The proportion of patients who underwent allogenic SCT was equivalent in the three treatment groups, and the outcome was similar as well. The 5-year overall survival rates were 34%, 34%, and 31%, respectively. In patients who reached CR and did not have a donor, autologous SCT was performed in 41%, 47% and 54% of patients in the mitoxantrone, idarubicin and daunorubicin arms, respectively. However, the disease-free survival (DFS) and survival from CR were significantly longer in the mitoxantrone and idarubicin arms than in the daunorubicin arm: the 5-year DFS rates were 37%, 37%, and 29%, respectively. So, in adult patients with AML who should not receive allogenic stem cell transplantation, the long-term efficacy of chemotherapy is enhanced through the use of mitoxantrone or idarubicin instead of daunorubicin.¹

This trial set a new standard of treatment for induction/consolidation in AML, with replacement of daunorubicin by either idarubicin or mitoxantrone.

This fruitful collaboration between the LG and GIMEMA began in 1986 with the AML-8 trial, with 1519 inclusions. In the AML-8A trial (testing autograft in first CR) in patients ≤45 years old, who reached complete remission, in those without a HLA donor, autograft provided longer disease-free survival than a second course of consolidation, whereas the best outcome was observed in patients with an HLA identical sibling, who had to be allografted.² In the last EORTC–GIMEMA AML-12 study, efficacy and toxicity of high dose vs standard dose of cytosine–arabinoside during induction and of IL-2 post-consolidation/autologous SCT were tested (2109 inclusions, data base was locked in August 2011 for final analysis).

With the development of molecular diagnosis permitting targeted therapy (for example *flt3* duplication/mutation), the era of large multicenter trials is probably over for AML. Intergroup trials now take on even more importance due to the small number of AML patients with particular molecular abnormality able to be enrolled by each group. One of the first examples is the CALGB/EORTC 06071 midostaurin trial in patients with *flt3* duplication/mutation.

3. Optimizing treatment in myelodysplastic syndrome

Medical management of MDS remains challenging. Over the past decade, epigenetic changes such as alterations in DNA methylation and histone modifications have been well described in MDS and are now recognized as therapeutic targets (epigenetic therapy). Since epigenetic silencing is reversible by pharmacological means, treatment approaches to target the aberrant epigenome of malignant cells have been developed. Building on encouraging phase II data with low-dose 5-aza-2'-deoxycytidine (decitabine, Dacogen) which resulted in a ~50% overall response rate in mostly higher-risk MDS patients, even in the presence of poor risk cytogenetics including complex karyotypes,³⁻⁵ the EORTC LG together with the German MDS Study Group conducted a phase III trial investigating the efficacy and toxicity of decitabine given intravenously over three days, repeated every six weeks.⁶

The control arm chosen for this trial was best supportive care (BSC), without the option of a "cross-over". The patient population was defined by higher-risk features such as blast cells excess of 11-30% in the bone marrow, and/or adverse cytogenetic abnormalities. To be registered for this trial of non-intensive treatment, patients were to be deemed ineligible for intensive chemotherapy by the treating physician. Between October 2002 and May 2007, 233 patients from 40 centers in nine European countries were randomized. According to the IPSS international score, 92% of patients had "higher-risk" disease and 32% fulfilled the WHO criteria of AML, i.e., at least 20% blasts. Notably, 53% of patients with known cytogenetic had poor-risk karyotypes. The median duration of MDS at randomization was only three months. The median number of decitabine courses administered was four, i.e., approximately six months of treatment. Median time for patients to exit from the study was 5.9 months in the decitabine arm *versus* 3.7 months in the BSC arm ($P < 0.001$).

Overall survival (OS), the primary end point of the study, was 10.1 months (median) in the decitabine arm *versus* 8.5 months in the BSC arm (HR=0.88; 95% CI: 0.66-1.17) and did not reach statistical significance.

Progression-free survival (PFS) was significantly prolonged in patients receiving decitabine ($P=0.004$), more than doubling of the median PFS (from 3 to 6.6 months) and a reduction of the hazard rate of 32% (HR=0.68; 95% CI: 0.52-0.88). The cumulative incidence of AML was significantly decreased with decitabine treatment compared to BSC (22% *versus* 33% at one year, 30% *versus* 43% at two years), with similar cumulative incidences of death without AML in both arms. There were 19% complete or partial remissions in the decitabine arm *versus* 0% in the BSC arm; hematologic improvement: 15% *versus* 2%. By multivariate analyses, IPSS high-risk, poor cytogenetic, <3 months of MDS duration and performance status ECOG 1 or 2 were independent prognostic factors for OS, PFS, and AML-free survival. Grade 3 to 4 infections were the most frequent adverse events in both arms. Quality of life was significantly improved in patients on the decitabine arm (self-reported fatigue and physical functioning), with borderline improvement of global health status. For most other quality of life scales, the trend was also in favor of decitabine.

This phase III trial of decitabine *versus* best supportive care in older, medically non-fit MDS patients with higher-risk MDS confirms, at a large scale, the efficacy of the drug in this patient population.⁷ Response rates were in line with previous trials with this drug performed previously by the principal investigators of this trial³⁻⁵: a significant prolongation of PFS and a reduction in the AML transformation rate. The lack of significant prolongation of OS may be due in part to the high proportion of patients with rapidly evolving high-risk MDS (AML?) having been recruited on this study. Further studies with decitabine are warranted in this patient population, e.g. comparison with the 4-week decitabine schedule, combinations with histone deacetylase inhibitors, and maintenance treatment with even lower-dose schedules.

4. Translational research: TET2 in MDS and AML

Until now the genes affected in MDS have remained largely unknown, despite the numerous chromosomal aberrations described in MDS. To identify relevant genetic lesions involved in the pathogenesis of MDS, J. Jansen and his colleagues in the EORTC NOCI (Network of Core Institutions) center of Nijmegen conducted SNP array-based genomic profiling and genomic sequencing in 102 patients with MDS and identified acquired abnormalities of the TET2 gene (deletions, missense or nonsense mutations) in 26% of the cases⁸ and in EZH2 in 5-10% of the patients.⁹ TET2 mutations were detected in most of the bone marrow cells (96%), including CD34+ progenitor cells, suggesting that TET2 mutations could be an early event during disease evolution. In normal bone marrow,

TET2 expression was elevated in granulocytes, suggesting a role in myelopoiesis. Until now, TET2 is the most frequently mutated gene in MDS.

The role of TET genes begins to unfold: the family of the three TET (TET1, TET2, TET3) proteins share two conserved regions that may be implicated in DNA epigenetic modification,¹⁰ and these regions are implicated in a novel form of DNA modification: conversion of 5-methylcytosine to 5-hydroxymethylcytosine. TET expression might lead to demethylation of DNA and de-repression of gene expression, and on the other hand TET mutation might lead to DNA hypermethylation and repression of tumor suppressor genes.

Translational research performed by the LG focused also on the prognostic impact of mutations and correlation with known genetic defects of this novel oncogene/tumor suppressor in *de novo* acute myeloid leukemia patients treated in the AML-12 trial (phase III EORTC trial 06991). This project, presented by S. Langemeijer and J. Jansen PhD, was granted by the EORTC in 2009 and results will be available soon from samples from four main EORTC centers and several GIMEMA centers. In MDS, our aim is to investigate the clinical response to demethylating agents in TET2 mutated versus TET2 wild-type MDS patients as well as in MDS with high versus low TET2 expression (non-mutated).

5. Strategy for the coming years

The molecular events driving the development of leukemia and myelodysplasia (core binding factors translocation, *flt-3* mutation, TET2 mutation, nucleophosmin mutation, etc.) are now systematically analyzed at diagnosis in AML and MDS, and some of these molecular markers are also used for monitoring residual disease in blood and bone marrow. According to the level of mutated allele, or to the presence of mutated oncogenes, the treatment of AML/MDS will be adapted. Now it is possible to get the results of the major mutation/translocation events within three working days which allows the inclusion of *de novo* untreated AML in randomized trials to test the efficacy of specific inhibitors, such as the first generation anti-*flt3* inhibitor midostaurin associated with "classical" chemotherapy (trial CALGB/EORTC 06071) [*flt3* mutation status performed in the NOCI center of Nijmegen (J. Jansen)]. The era of large trials including more than 2000 AML patients (such as EORTC trials AML-12 and AML-10) is now finished, but development of intergroup trials focusing on subgroups of AML bearing specific genetic abnormalities is now mandatory to validate the "targeted approach" of driving molecular events. Addition of promising drug in AML like clofarabine to standard chemotherapy is currently tested in a phase II trial in EORTC and GIMEMA centers (AML14A, 06061).

The treatment of elderly AML, where complex karyotypes are more frequent, is a challenge for the LG. As have others, we noted the absence of improvement in OS (<10% at 5 years) for patients included in our subsequent elderly AML trials. For elderly patients with AML, unfit for intensive chemotherapy, our EORTC-GIMEMA trial (AML-19) is currently testing the use of very low dose of gemtuzumab ozogamycin to improve quality of life and overall survival. We are planning to build a platform for patients over 55 years of age, including systematic tumor cell banking at diagnosis, a standard 3+7 treatment (versus experimental "pick the winner" arm), followed by reduced intensity conditioning regimen (RIC) allograft in patient in remission, provided performance status is adequate, and there are available donors. The adoptive immunotherapy of this allograft represents a very interesting option in these patients, who have a very high rate of relapse within one year.

Concerning high-risk MDS, the next step will be the improvement of the results obtained with demethylating agents (decitabine and 5-azacytidine). The duration of hematological responses with demethylating agents is limited, and promising preliminary results were obtained when these agents are used in combination with HDAC inhibitors, such as vorinostat.¹¹

6. Conflict of interest statement

Stefan Suciu, Matthias Karrasch, Giovanna Meloni, Sergio Amadori, Michael Lübbert, Roel Willemze, Petra Muus, Ann Hagemeijer, Dominique Bron, Joop Jansen, and Pierre Wijermans declare no conflicts of interest. Boris Labar consulted for Allexion, received honoraria from Pfizer, and received honoraria and travel reimbursement from Novartis. Theo de Witte consulted for and received honoraria and research funds from Novartis, consulted for and received honoraria from Clavis and Celgene, and received research funds from Johnson & Johnson. Jean-Pierre Marie received travel reimbursement from GSK. Dominik Selleslag consulted for and received honoraria and research funds from Amgen, Novartis, Celgene, MSD, Pfizer, Sunesis, and GSK, and consulted for and received honoraria from Shire and Mundipharma. Frédéric Baron received honoraria, research funds and travel reimbursement from Genzyme, research funds and travel reimbursement from Amgen, and travel reimbursement from Novartis, BMS, and Roche.

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