Treatment of pediatric acute lymphoblastic leukemia

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Acute lymphoblastic leukemia (ALL) is the most common type of cancer in children. Although it may affect children of any age, there is a peak modal distribution between 3 and 6 years. The presenting features may be quite variable but in most case the diagnosis is promptly reached by morphological examination of a bone marrow aspirate.

Recent advances in the treatment of childhood ALL may be regarded as a paradigm of the effectiveness of medical science in the management of formerly incurable diseases. Starting from the middle of the last century, optimal use of the few antileukemic agents already available at that time, together with improved skills in patient stratification and better supportive therapy produced a steady improvement in treatment outcome, so that the current cure rate is now about 80%.[1,2]

Identification of patients with different risks of an adverse prognosis

In order to provide treatment that is appropriate for the individual risk of treatment failure, i.e. less aggressive in patients at lower risk and more aggressive in patients at higher risk, presenting features, such as age and leukocyte count, have been traditionally considered. These features have been used to define standard, intermediate, and high risk groups of patients. Patients with B-cell precursor ALL, an age between 1 and 9 years old, and a leukocyte count $<50 \times 10^9$/L, usually belong to the standard-risk group.[3] Additional factors such as race or immunophenotype, have also been widely considered. Yet, as expected, most of the prognostic factors appear to be strictly dependent on the efficacy of the treatment applied. Thus, for mature B-cell leukemia lost its adverse prognostic value many years ago and the risk of treatment failure in T-cell ALL is now also comparable to that in case of ALL B-cell precursor as BCP cases.

Clonal genetic abnormalities

It has been known for many years that certain genetic abnormalities are associated with different prognoses.[4,5] About one half of children with ALL have B-cell precursor ALL, more than 50 chromosomes (hyperdiploidy), and translocation t(12;21) leading to the *TEL-AML1* fusion gene; such children are generally considered to have a favorable prognosis in most contemporary therapeutic settings,[6] although this belief has been questioned by some groups. About one half of patients with T-ALL harbor mutations of *NOTCH1*[7] but the prognostic relevance of this has not yet been clarified.

The Pediatric Oncology Group, as well as other groups, reported that a blast DNA index between 1.16 and 1.6 is associated with a favorable prognosis. Trisomies 4, 10, and 17 also confer a better prognosis.[8-10]

In contrast, patients with fewer than 45 chromosomes (hypodiploidy), which occur in no more than 2% of cases, have a dismal prognosis. The translocation t(4;11) and the *MLL-AF4* fusion gene are observed in about one half of children younger than 1 year old; these infants have a very poor prognosis. The translocation t(9;22) and the *BCR-ABL* fusion are common in adults but present in only 2.5% of cases of childhood ALL; the prognosis of these children is not uniform, being very poor in some of them, particularly those aged 10 years or more, and with a leukocyte count of $>50 \times 10^9$/L.[11]

Factors influencing drug disposal in the host

Several drugs have been the focus of pharmacokinetic studies, aimed at targeting the plasma levels in the hope of providing optimal individual levels to each patient. The St. Jude Children's Research Hospital in Memphis (USA) has been a pioneer in this field.[12] Their studies suggested that antimetabolites may have different effects in patients with specific genetic polymorphisms due to rapid clearance or inactivation, affecting the treatment outcome. However, the ultimate prognostic relevance of such genetic polymorphisms may be hard to predict becausse of the use of combination therapy.

In vivo *response to therapy*

It is easy to expect that a patient who responds to antileukemic therapy could have a better prognosis. Yet, having this information after completion of treatment is of little help for the leukemia specialist who must choose the treatment and possibly minimize its toxicity.

The Berlin-Frankfurt-Münster (BFM) co-operative group introduced the concept of an initial window of treatment with prednisone, escalating from 20 to 60 mg/m^2, and one age-adjusted dose of intrathecal methotrexate. This provided the opportunity not only to have a milder start of cytoreduction, particularly useful in patients with a very high leukocyte count, but also to test the *in vivo* sensitivity of leukemic blasts to steroid-induced apoptosis. Patients with more rapid blast clearance in the peripheral blood (e.g. $<1 \times 10^9$ blasts/L on day 8) were defined as *prednisone-good-responders*, and definitely had a more favorable outcome, while the remaining 10% patients (*prednisone-poor-responders*) had a higher probability of early or late treatment failure.[13] This concept has been widely used in clinical practice to stratify patients by all groups using BFM-type chemotherapy. As reported in this issue of the journal, this concept was extended by the Tokyo Children's Cancer Study Group (TCCSG), with the finding that patients with no blasts in the peripheral blood on day 8, who accounted for one third of all

the cases, had an excellent outcome with over 90% surviving at 4 years.[14]

One main next step for evaluation is the achievement of morphological remission by the end of induction therapy, usually between day 28 and 42. It is well known that patients failing to have <5% blasts in the bone marrow, as assessed by morphology, have a dismal prognosis, although many of them may have a chance to achieve morphological remission by subsequent chemotherapy, thus opening the way to alternative modalities of consolidation.

During the last decade, measurement of blast reduction during the first weeks became a relevant issue. Morphological evaluation is not sufficiently accurate for this purpose and methods aimed at detecting of limited numbers of blast cells (*minimal residual disease*) in the bone marrow, or even in the peripheral blood, have been explored. Such methods may provide the advantage of an early information allowing significant modifications of the treatment schedule according to the prognostic indications. Both flow cytometry and polymerase chain reaction (PCR) analysis have been used for the detection of minimal residual disease. Patients with leukemic blast concentrations below 0.01% by the end of induction therapy have a good prognosis because of their low risk of relapse. Contrariwise, when the concentration of leukemic blasts is 10% on day 15, or 1% by the end of induction therapy or later on, the risk of leukemia relapse is very high.[15-16]

PCR analysis is a very sensitive method for identifying very low risk patients, although it is very expensive and time-consuming, and up to 20% of patients may be ineligible for technical reasons by the more stringent protocols of analysis. Its extensive use in the AIEOP-BFM-ALL 2000 stydy recently provided evidence that PCR analysis can discriminate patients with different prognoses even within groups defined as non-high risk by traditional criteria.[17] Flow cytometry appears a very promising technique for the detection of minimal residual disease, is relatively cheap and almost all patients can be studied within hours. Its extensive clinical use by the St. Jude Children's Research Hospital in Memphis demonstrated its feasibility in a single, large institution. More recently, two large co-operative studies have confirmed the feasibility of this method in children with ALL.[18-20]

Front-line treatment of childhood acute lymphoblastic leukemia

With the only exception of patients with mature B-cell ALL, for whom a specific short and very intense chemotherapy program has been developed by most groups, all patients with childhood ALL are treated according to a similar strategy. Front-line therapy consists of several phases, with different aims; initial *induction* (of the first remission) therapy is delivered over 4-5 weeks; central nervous system (CNS)-directed therapy is administered to prevent meningeal progression or relapse; intensive post-remission chemotherapy is aimed at reducing residual leukemia; finally continuation therapy provides further reduction of the risk of relapse.

Induction therapy

Most leukemia specialists consider that initial cytoreduction in the range of two-logs, leaving a residual blast count of less than 5% by the end of induction therapy, is crucial for a final cure. To achieve this goal, and thus restore normal hematopoiesis and decrease the risk of infection, a combination of three to five drugs is delivered over 4-5 weeks. All of the current regimens include weekly vincristine and daily steroids. Yet, despite their wide and long-lasting use, there is still great debate on the optimal type and dosage of steroids. Traditionally, oral prednisone at a dose of 40 mg/m^2 for 28 days has been considered the standard by many groups. Yet, we now have clear evidence that more aggressive steroid therapy provides stronger leukemia control. The recent introduction of dexamethasone in several co-operative studies has produced a significant improvement in leukemia control, both by enhancing the coverage of the so-called extramedullary spaces (including CNS and testicles) and by preventing subsequent relapse.[20,21,22] The use of high-dose dexamethasone is, however associated with a higher rate of morbidity, including bacterial and fungal infections, behavioral alterations, and avascular necrosis. The benefits of its use must, therefore, be balanced against the risks of toxic death and high rates of severe morbidity. At present, whether it is better to use higher doses of prednisone[23] or dexamethasone, at 6 or 10 mg/m^2, remains the object of discussion by many groups.

The addition of a third or fourth drug, such as asparaginase or an anthracycline, may not be necessary in standard risk patients but is definitely of help in patients at higher risk. Using this strategy, modern trials are able to bring up to 98% of children with ALL into morphological remission. Whether less intensive induction therapy may be balanced by a more aggressive subsequent approach has been questioned by some studies.[24]

Prophylaxis of meningeal leukemia

Starting from the middle of the last century, the pioneering activity of D. Pinkel at the St. Jude Children's Research Hospital provided insights into the potential of radiotherapy to decrease the treatment failure frequently occurring as a consequence of CNS dissemination in patients during apparent control of disease in the bone marrow The benefit of radiotherapy is, however outweighed by the occurrence of secondary neoplasms – especially CNS tumors in the irradiated field – endocrine and growth disorders, and neurocognitive dysfunction. St. Jude Children's Research Hospital documented that previous irradiation in subjects cured from childhood ALL was associated with a 20.9% cumulative risk of second neoplasms at 30 years, together with higher mortality and unemployment rates than those in the general population.[25]

Nowadays, continuous improvement in the efficacy of chemotherapy has allowed irradiation for CNS prophylaxis to be replaced by a wider use of dexamethasone, high-dose methotrexate and extended intrathecal chemotherapy.[26]

Methotrexate at medium to high doses is frequently

used during this phase because of its ability to diffuse in the CNS. It has been documented that high-dose methotrexate improves the outcome in patients with T-cell ALL, because of a lower accumulation of the active polyglutamated metabolite in T-cell ALL than in B-cell precursor ALL. Comparably, patients with *TEL-AML1* or *E2A-PBX1* may also benefit from higher doses of methotrexate.

High-dose methotrexate is not, however, completely free from toxicity. Patients receiving higher doses and more courses of intravenous metrotrexate were found to be at a higher risk of leukoencephalopathy; although many of the cerebral changes resolved after completion of therapy, their effects on neurocognitive function and quality of life in survivors remain to be determined.[27]

Cranial irradiation is still indicated for a small minority of patients from selected subgroups, such as those with T-cell ALL presenting with leukocyte counts >100×10⁹/L,28 or CNS disease, or failure to achieve remission. The hypothesis of reducing the radiation dose to 12 Gy for prophylaxis and to 18 Gy for those with CNS disease is still supported only by limited experience, mainly from the BFM group.

Intrathecal chemotherapy with methotrexate is thus, at present, the standard for all patients, at least during the first 6-12 months of treatment. The duration of intrathecal therapy should also be adjusted depending on the use of radiotherapy and some high-dose agents. Careful attention should be paid to the prevention of traumatic lumbar punctures, especially at diagnosis, when patients may have abundant circulating blasts.[27]

In parallel, irradiation has been used for treatment of testicular leukemia. At present, some institutions suggest that it may be omitted provided effective systemic chemotherapy is given.[29]

Late intensification or reinduction

It is a commonly accepted concept that patients in apparent complete remission still have minimal residual disease. Thus, exposure to intensification therapy may provide additional and possibly final leukemia control. To this purpose, many groups utilize a treatment program that includes drugs identical or very similar to those used in induction. The value of late intensification or *reinduction* was strongly suggested by early experience of the BFM group and later confirmed by other groups.[2,4,10,24,30-32] Whether this benefit comes from re-exposure to vincristine and prednisone was questioned by the Children's Cancer Group.[24]

Exposure to asparaginase as post-induction intensification therapy provided excellent results with low morbidity, including low rates of glucose intolerance and thrombosis. Several forms of asparaginase are commercially available; since their pharmacokinetic profiles differ, the ideal dose and frequency of administration of asparaginases may vary. This has produced some problems in the interpretation of the results of several randomized trials addressing the role of asparaginase during late intensification.[33,34]

Higher cumulative dose of steroids in adolescents have been repeatedly associated with a risk of avascular necrosis, thus suggesting that these drugs should be administered in alternate weeks to reduce this complication, which necessitates hip replacement in most patients.[35]

Allogeneic hematopoietic stem cell transplantation

Despite recent improvements, some patients with childhood ALL fail to benefit from modern chemotherapy. Thus patients who do not achieve remission or have a leukemia relapse, especially when this occurs during or soon after treatment completion, have been addressed to allogeneic hematopoietic stem cell transplantation as an ultimate form of treatment intensification, associated also with the benefit of *immunotherapy* provided by the donor immunity.

At present, co-operative efforts have provided large series of patients evaluable for comparative analysis of outcome of front-line treatment based on chemotherapy with or without transplantation. Nevertheless, such comparisons are often biased by patient selection and the variable of time to transplantation. In an international prospective study comparing transplantation strategies allocated by genetic randomization, children with very high-risk ALL benefited more from related-donor transplantation than from chemotherapy, with a gap between the two strategies which increased as the risk profile of the patient worsened.[36]

In the subset of patients with *BCR-ABL*-positive ALL treated during the 1980s and 1990s, transplantation of bone marrow from an HLA-matched related donor was superior to other types of transplantation, and to intensive chemotherapy alone, in prolonging initial complete remissions.[11] Whether it is beneficial for another rare subset of ALL patients with a dismal outcome, i.e. infants with t(4;11), remains controversial.[37] Continuous improvements in the techniques of unrelated donor selection make this strategy more appealing for a larger proportion of children with refractory, as well as relapsed, ALL.[38] Thus, the indications for transplantation should be continuously evaluated in the light of improvements in this procedure and in chemotherapy.

Continuation therapy

The real mechanism underlying the protective effect of continuation therapy on childhood ALL remains unclear. Whenever treatment duration was shortened to 12 to 18 months, inferior results were obtained. Thus, although we have now learned that up to two-thirds of patients may be cured with only 12 months of treatment,[39] since we are not yet able to recognize them upfront, the duration of treatment in most current trials is still at least 24 months.[40,41]

The most popular combination is daily mercaptopurine and weekly methotrexate, orally. It is common experience that moderate cytopenia and rises in the concentrations of liver enzymes are promptly reversible, herald intracellular accumulation of active metabolites, and are associated with a lower risk of relapse. Mercaptopurine is more effective when administered in the evening, and should not be given with milk or milk products, since xanthine oxidase can degrade the drug.[40] Patients with inherited thiopurine-

S-methyltransferase deficiency show cytopenia and may require a lower dose of mercaptopurine.[41] Intramuscular administration of methotrexate may circumvent poor compliance in adolescents.

In an attempt to intensify this treatment component, dexamethasone and vicnristine pulses have been administered but with no advantage.[42]

A potential protective role of extended intrathecal methotrexate during continuation therapy is likely blurred when patients have been treated with intensive chemotherapy schedules.

Future directions

Given the high cure rate achieved in contemporary trials, special attention must be paid to the quality of life of cured children. Thus, many groups are limiting the use of anthracyclines and alkylating agents in standard-risk patients. Topo-II inhibitors, such as etoposide, have been associated with a risk of secondary myeloproliferation and their use is, therefore, currently restricted to very high-risk patients. Patients with associated genetic conditions may need special attention;[43] for example, patients with Down's syndrome require reduction of the dose of methotrexate, and those with chromosomal instability syndromes are usually excessively sensitive to alkylating agents and irradiation.

The use of imatinib mesylate is currently under investigation in patients with childhood *BCR/ABL*-positive ALL; if its benefit is confirmed, second-generation ABL kinase inhibitors may enter clinical practice.

A pegylated form of asparaginase is currently being tested by several groups in order to reduce the number of administrations and possibly to increase the efficacy of the asparaginase. Liposomal encapsulated drugs, such as anthracyclines, are also being evaluated for their possible lower toxicity and better diffusion, but have not, so far reached most frontline schedules. Clofarabine, a deoxyadenosine analog, was recently granted approval for use in children with advanced ALL, thus being the only anticancer drug to receive a primary indication for use in children over the past decade. Ongoing studies are exploring the benefit of its use in combinations in refractory very high risk patients but also as front-line therapy.[44]

Finally, experience gained in the management of childhood ALL is being transferred not only to adolescents, traditionally also cared for in adult hematology units, but also to young adults, in whom strategies used for childhood ALL have already proven to be largely of benefit, although not in all settings.[45,46]

Monoclonal antibodies against specific epitopes of ALL blasts have been repeatedly tested but, thus far, none has become widely used in childhood ALL. Finally, studies of gene expression profiles and microRNA, are expected to improve the classification of ALL and possibly indicate treatment modifications.

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