ELSEVIER
SAUNDERS

PEDIATRIC CLINICS
OF NORTH AMERICA

Pediatr Clin N Am 55 (2008) 1–20

Biology and Treatment of Acute Lymphoblastic Leukemia

Rob Pieters, MD, MSc, PhD[a,*],
William L. Carroll, MD[b]

[a]*Department of Pediatric Oncology and Hematology, Erasmus MC-Sophia Children's Hospital, Dr Molewaterplein 60, 3015GJ Rotterdam, The Netherlands*
[b]*Division of Pediatric Hematology/Oncology, New York University Medical Center, 160 East 32nd Street, 2nd Floor, New York, NY 10016, USA*

Acute lymphoblastic leukemia (ALL), the most common type of cancer in children, is a heterogeneous disease in which many genetic lesions result in the development of multiple biologic subtypes. The etiology of ALL is characterized by the acquisition of multiple consecutive genetic alterations in the (pre)leukemic cells. In the most common genetic subtypes of ALL, the first hit occurs in utero [1], as evidenced, for example, by the presence of the *TEL/AML1* gene fusion or hyperdiploidy in neonatal blood spots on Guthrie cards. These first genetic abnormalities are, in fact, initiating preleukemic cells, not leukemic ones, because most children whose neonatal blood spots show a genetic defect typically associated with leukemia never develop leukemia. Also, such preleukemic cells harbor additional genetic abnormalities. T-cell acute lymphoblastic leukemia (T-ALL) is an exception, because the majority of genetic lesions described in T-ALL seem not to occur in the neonatal blood spots [2].

Today, with intensive multiagent chemotherapy, most children who have ALL are cured. The factors that account for the dramatic improvement in survival during the past 40 years include the identification of effective drugs and combination chemotherapy through large, randomized clinical trials, the recognition of sanctuary sites and the integration of presymptomatic central nervous system (CNS) prophylaxis, intensification of treatment using existing drugs, and risk-based stratification of treatment. The many national or institutional ALL therapy protocols in use tend to stratify patients in a multitude of different ways. Treatment results often are not published for the

* Corresponding author.
E-mail address: rob.pieters@erasmusmc.nl (R. Pieters).

0031-3955/08/$ - see front matter
doi:10.1016/j.pcl.2007.11.002

overall patient group but rather are reported only for selected subsets of patients. This limitation hampers the comparison of outcomes in protocols. In 2000, the results of ALL trials run in the early 1990s by the major study groups were presented in a uniform way [3–12]. The 5-year event-free survival (EFS) rates seemed not to vary widely, ranging from 71% to 83% (Table 1). Overall remission rates usually were 98% or higher.

Risk-based stratification allows the tailoring of treatment according to the predicted risk of relapse. Children who have high-risk features receive aggressive treatment to prevent disease recurrence, and patients who have a good prognosis receive effective therapy but are not exposed to unnecessary treatment with associated short- and long-term side effects. Clinical factors that predict outcome and are used for stratification of patients into treatment arms are age, gender, and white blood cell count at presentation. Biologic factors with prognostic value are the immunophenotype and genotype of the leukemia cells. Another predictive factor is the rapidity of response to early therapy, such as the decrease in peripheral blood blast count in response to a week of prednisone or the decrease in bone marrow blasts after 1 to 3 weeks of multiagent chemotherapy. More recently the determination of minimal residual disease (MRD) in the bone marrow during the first months of therapy using flow cytometry or molecular techniques has been shown to have a high prognostic value and therefore is used for stratification in many contemporary trials. The detection of MRD accurately distinguishes very good responders to therapy from those who will

Table 1
Treatment results from major clinical trials in childhood acute lymphoblastic leukemia conducted in the early 1990s

Study group	Years of study	Patient number	Overall 5-year event-free survival (%)	B-lineage ALL 5-year event-free survival (%)	T-lineage ALL 5-year event-free survival (%)
DFCI-91-01	1991–1995	377	83	84	79
BFM-90	1990–1995	2178	78	80	61
NOPHO-ALL92	1992–1998	1143	78	79	61
COALL-92	1992–1997	538	77	78	71
SJCRH-13A	1991–1994	167	77	80	61
CCG-1800	1989–1995	5121	75	75	73
DCOG-ALL8	1991–1996	467	73	73	71
EORTC-58881	1989–1998	2065	71	72	64
AIEOP-91	1991–1995	1194	71	75	40
UKALL-XI	1990–1997	2090	63	63	59

Abbreviations: AIEOP, Associazione Italiana Ematologia Oncologia Pediatrica; BMF, Berlin-Frankfurt-Münster; CCG, Children's Cancer Group; COALL, Co-operative Study Group of Childhood Acute Lymphoblastic Leukemia; DCOG, Dutch Childhood Oncology Group; DFCI, Dana Farber Cancer Institute; EORTC-CLG European Organization for the Research and Treatment of Cancer; NOPHO, Nordic Society of Pediatric Haematology and Oncology; SJCHR, St. Jude Children's Research Hospital; UKALL United Kingdom Acute Lymphoblastic Leukemia.

respond poorly to therapy, irrespective of the biologic subtype of ALL and the underlying mechanism of this response [13]. In several protocols, MRD is used to stratify patients for reduction of therapy (ie, patients who are MRD negative especially at early time points) or intensification of therapy (ie, patients who are MRD positive at later time points).

Age and immunophenotype

Over the years, age has remained an independent predictor of outcome (Table 2). Children aged 1 to 9 years have the best outcome; children and adolescents aged 10 to 20 years have a slightly worse outcome, which is associated in part with a higher incidence of T-cell leukemia and a lower incidence of favorable genetic abnormalities such as *TEL/AML1* and hyperdiploidy. For adults, survival rates decrease further with increasing age. When results are corrected for differences in immunophenotype, ALL cells from older children and adults are more resistant to multiple antileukemic drugs than are cells from children in the first decade of life [14,15].

Infants diagnosed at less than 1 year of age have a relatively poor outcome that is associated with a high incidence of the unfavorable very immature proB-ALL phenotype and especially the presence of *MLL* gene rearrangements [16]. The poor outcome has led physicians in the United States, Japan, and the International Interfant collaborative group including European and non-European countries and institutes to develop specific protocols to treat infant ALL [13,17,18]. Biologic characteristics of infant ALL cells are described later in the paragraph discussing the *MLL* gene.

T-cell ALL is detected in approximately 15% of childhood ALL. It is characterized by a relative resistance to different classes of drugs when compared with B-lineage ALL [14]. T-cell ALL cells accumulate less methotrexate polyglutamates and less cytarabine triphosphate than precursor B-ALL cells [19]. With risk-adapted therapy the outcome of T-cell ALL now approaches that of B-lineage ALL in many study groups (see Table 1).

Approximately 85% of childhood ALL is of B lineage, mainly common or preB ALL. A very immature subtype characterized by the lack of CD10

Table 2
Clinical and biologic factors predicting clinical outcome

Factor	Favorable	Unfavorable
Age at diagnosis	1–9 years	$<$ 1 or $>$ 9 years
Sex	Female	Male
White blood cell count	Low (eg, $<$ 50 or $<$ 25 $\times$ 10^e9/L)	High (eg, $>$ 50 or $>$ 25 $\times$ 10^e9/L)
Genotype	Hyperdiploidy ($>$ 50 chromosomes) t(12;21) or *TEL/AML1* fusion	Hypodiploidy ($<$ 45 chromosomes) t(9;22) or *BCR/ABL* fusion t(4;11) or *MLL/AF4* fusion
Immunophenotype	Common, preB	ProB, T-lineage

expression (proB ALL) is associated with a high incidence of *MLL* gene rearrangements and an unfavorable outcome. Mature B-lineage ALL, defined by the presence of immunoglobulins on the cell surface, has a favorable outcome only when treated with B-non-Hodgkin lymphoma protocols.

Genetics

Hyperdiploidy

Hyperdiploidy (a DNA index > 1.16 or > 50 chromosomes per leukemia cell) is found in approximately 25% of children who have B-lineage ALL. It is associated with a favorable outcome, especially when extra copies of chromosome 4, 10 or 17 are present [20]. Hyperdiploid ALL cells have an increased tendency to undergo apoptosis, accumulate high amounts of methotrexate polyglutamates, and are highly sensitive to antimetabolites and L-asparaginase [21].

TEL/AML1

The *TEL/AML1* fusion, also found in approximately 25% of cases, is mutually exclusive with hyperdiploidy and also is associated with a favorable outcome. It is formed by a fusion of the *TEL* gene on chromosome 12 encoding for a nuclear phosphoprotein of the ETS family of transcription factors and the *AML1* gene on chromosome 21, a transcription factor gene encoding for part of the core-binding factor. The *TEL/AML1* fusion probably inhibits the transcription activity of the normal *AML1* gene involved in proliferation and differentiation of hematopoietic cells. *TEL/AML1* fusion is associated with a high chemosensitivity, especially for L-asparaginase [22]. The mechanism behind this asparaginase sensitivity remains unclear but is not caused by a low asparagines synthetase activity in the leukemic cells [23,24]. *TEL/AML1*-rearranged cells also may be more sensitive to other drugs, especially anthracyclines and etoposide [25].

Both hyperdiploidy and *TEL/AML1* occur mainly in children younger than 10 years of age with common/preB ALL and are rare above this age and in other ALL immunophenotypes.

MLL

Abnormalities of the mixed lineage leukemia (*MLL*) gene on chromosome 11q23 occur in only approximately 2% of children above the age of 1 year, although it is present in approximately 80% of infants who have ALL. All types of *MLL* gene rearrangements, such as *MLL/AF*4 created by t(4;11), *MLL/ENL* created by t(11;19), and *MLL/AF9* created by t(9;11), are associated with a poor outcome in infants who have ALL [17]; in older children this poor outcome may only hold true for the presence of *MLL/AF4* [26]. The *MLL/AF9* rearrangement occurs in older infants

and is characterized by a more mature pattern of immunoglobulin gene rearrangements, suggesting another pathogenesis [17,27].

The precise actions of the fusion products involving *MLL* are not known, but they are associated with abnormal expression of *HOX* genes, which may lead to abnormal growth of hematopoietic stem cells [28]. ALL cells with *MLL* gene abnormalities are highly resistant to glucocorticoids in vitro and in vivo and also to L-asparaginase [14,17,29]. These cells, however, show a marked sensitivity to the nucleoside analogues cytarabine and cladribine [30]. This sensitivity is related to a high expression of the membrane nucleoside transporter ENT1 [31]. *MLL*-rearranged ALL cells do not show a defective methotrexate polyglutamation [32] and have no overexpression of multidrug resistance proteins [33]. Methotrexate pharmacokinetics might be different in the youngest infants [34].

BCR-ABL

The translocation t(9;22) fuses the *BCR* gene on chromosome 22 to the *ABL* gene on chromosome 9 causing an abnormal *ABL* tyrosine kinase activity associated with increased proliferation and decreased apoptosis. The *BCR/ABL* fusion is found mainly in common and preB ALL. The incidence of *BCR/ABL* increases with age: it is seen in approximately 3% of children who have ALL but in approximately 25% of adults who have ALL. The presence of *BCR/ABL* predicts a poor outcome.

Children who have *BCR/ABL*-rearranged ALL or *MLL*-rearranged ALL more often show a poor response to prednisone [29,35] and have high levels of MRD after induction therapy.

Genetics in T-cell acute lymphoblastic leukemia

The prognostic value of genetic abnormalities in T-ALL is less clear [36]. Ectopic expression of *TAL-1* is caused by the translocation t(1;14) in only a few percent of T-ALL cases or, more often, by the *SIL-TAL* fusion transcript. Activation of *HOX11* by the translocations t(10;14) and t(7;10) occur in approximately 10% of T-ALL cases. Two recently described abnormalities occur frequently and exclusively in T-ALL. These are the ectopic expression of *HOX11L2*, mainly caused by the translocation t(5;14), in approximately 25% of T-ALL cases and activating mutations of the *NOTCH1* gene in 50% of T-ALL cases. *NOTCH1* mutations are not associated with a poor outcome and may be associated with a favorable outcome [37].

Others

Many other recurrent genetic and molecular genetic lesions exist in small subsets of childhood ALL such as the translocation t(1;19) leading to a *E2A-PBX1* fusion detected in less than 5% of precursor B-ALL, mainly preB ALL. Although in the past this translocation had been associated with

a poor prognosis, this is not longer true with contemporary treatment protocols. Two percent of precursor B-lineage ALL cases harbor an intrachromosomal amplification of chromosome 21 that is associated with poor survival [38]. Hypodiploidy (< 45 chromosomes) is detected in only 1% of children who have ALL and is associated with poor outcome, particularly in the low-hypodiploid (33–39 chromosomes) or near-haploid cases (23–29 chromosomes) as shown in a recent retrospective international study [39].

A discussion of all other abnormalities is beyond the scope of this article. It should be mentioned that children who have Down syndrome and ALL do not have a better outcome and perhaps even have a worse outcome than other ALL cases because they lack favorable genetic features [40,41].

Therapy

The backbone of contemporary multiagent chemotherapeutic regimens is formed by four elements: induction, CNS-directed treatment and consolidation, reinduction, and maintenance.

Induction

The goal of induction therapy is to induce morphologic remission and to restore normal hematopoiesis. Induction therapy contains at least three systemic drugs (ie, a glucocorticoid, vincristine, and L-asparaginase) and intrathecal therapy. The addition of an anthracycline as a fourth drug is matter of debate. In some protocols all patients receive an anthracycline; in other protocols it is reserved for high-risk cases. The induction phase aims to induce complete morphologic remission in 4 to 6 weeks.

Central nervous system–directed treatment and consolidation

CNS-directed therapy aims to prevent CNS relapses and to reduce the systemic minimal residual leukemia burden. CNS therapy usually is achieved by weekly or biweekly intrathecal therapy along with systemically administered drugs such as high-dose methotrexate (MTX) and 6-mercaptopurine (6-MP). Some groups rely on other drugs (eg, cyclophosphamide, cytarabine) in the consolidation phase to reduce systemic tumor burden further.

Reinduction

Reinduction therapy or delayed (re)intensification most often uses drugs comparable to those used during induction and consolidation therapy and has clearly shown its value by reducing the risk of relapse.

Maintenance

Therapy for ALL is completed by prolonged maintenance therapy for a total treatment duration of 2 years, or even longer in some protocols.

Maintenance consists of daily 6-MP and weekly MTX. In some protocols additional pulsed applications of a glucocorticoid and vincristine and intrathecal therapy are administered.

A fifth element, allogeneic stem cell transplantation (SCT), is reserved for only a small number of selected patients in first complete remission. The contribution of specific parts of treatment depends on the total therapy administered to a patient. A few important topics for which new data have been produced recently are discussed in the following sections.

Anthracyclines in induction?

It is unclear if addition of an anthracycline to a three-drug induction regimen is of benefit. Regimens that do not contain anthracycline are less myelosuppressive. Studies performed by the Children's Cancer Group, however, showed that selected patients younger than 10 years of age did not benefit from the addition of an anthracycline, whereas selected older children did [42].

Dexamethasone or prednisone?

Several recent randomized studies have shown that the substitution of prednisone (approximately 40 mg/m^2) by dexamethasone (approximately 6 mg/m^2) significantly decreases the risk of bone marrow and CNS relapses when used in what are thought to be equipotent dosages [43,44]. One Japanese study, however, did not confirm the advantage of using dexamethasone [45]. The benefit of dexamethasone may result from higher free plasma levels and a better CNS penetration or from the fact that the presumed equivalent antileukemic activity for prednisone/dexamethasone is not a 6:1 dose ratio but is higher, as some (but not all) in vitro experiments suggest [46,47]. At this dose ratio dexamethasone also results in more toxicity than prednisone [43]. In vitro, a strong cross-resistance to prednisone and dexamethasone exists in ALL cells.

Which dose intensity of which asparaginase?

Randomized studies have revealed that at the same dose schedules, the use of L-asparaginase derived from *Escherichia coli* resulted in significant better EFS and overall survival (OS) rates than when asparaginase derived from *Erwinia chrysanthemi* (Erwinase) was used [48,49]. This difference results from differences in the half-lives of the drugs, and the difference presumably would not be found if Erwinase were given in an adequate dose-intensity schedule. The dose-intensity schedule to achieve complete asparagine depletion is 5000 units/m^2 every 3 days for *E coli* asparaginase. Erwinase must be scheduled more frequently than *E coli* asparaginase to achieve the same asparagine depletion. For the pegylated type of *E coli* asparaginase (PEG-asparaginase), 2500 units/m^2 once every 2 weeks leads

to the same pharmacodynamic effects. Lower doses of PEG-asparaginase (1000 units/m^2) also lead to complete asparagine depletion in serum but not in the cerebrospinal fluid [50].

Intensification of asparaginase in induction and reinduction has improved outcomes in different studies [51–53]. Also, asparaginase intolerance was an important factor predicting an inferior outcome [54,55]. Allergic reactions usually are responsible for the discontinuation of asparaginase. Allergic reactions occur mainly when the drug is readministered in reinduction several weeks after first exposure during induction. In addition, the presence of asparaginase antibodies may lead to inactivation of the drug. Consequently, many investigators favor the use of the less immunogenic PEG-asparaginase from therapy outset rather than using it only after allergic reactions have occurred. In the light of these data, pharmacodynamic monitoring of asparaginase administration may prove very important for individual children who have ALL.

Which central nervous system–directed therapy?

To clarify the role of different CNS-directed therapies, a meta-analysis was published in 2003 [56]. From this analysis it became clear that long-term intrathecal therapy leads to EFS rates comparable with those of radiotherapy. Radiotherapy seemed to be more effective than high-dose MTX in preventing CNS relapse, but intravenous MTX reduced systemic relapses, resulting in comparable EFS rates for high-dose MTX and radiotherapy. It was concluded that radiotherapy can be replaced by multiple intrathecal doses of chemotherapy and that intravenous MTX reduces systemic relapses. It is still unclear whether intrathecal triple therapy (glucocorticoid, MTX, cytarabine) has any advantage over the use of intrathecal MTX as single drug. A recent Children's Cancer Group study suggested that intrathecal triple therapy prevented CNS relapse but did not improve OS because fewer bone marrow relapses occurred when intrathecal MTX was used as a single agent [57].

The results of CNS-directed therapy depend on the treatment used. For example, the use of systemic dexamethasone reduces the incidence of CNS relapse. The comparison of different CNS preventive regimens is hampered because results are described for heterogeneous groups of patients. In several protocols, radiotherapy is still given to selected groups of high-risk patients such as those who have T-ALL with high white cell counts and children who have CNS involvement at diagnosis. Cranial radiotherapy is specifically toxic for very young children because of its detrimental effect on cognitive function.

What type of reinduction/intensification and maintenance?

Maintenance therapy consists of daily oral 6-MP and weekly intravenous or oral MTX. The intravenous administration of MTX may overcome compliance problems, but there is no evidence that it is more effective than oral

MTX. Several randomized studies have shown that the use of thioguanine offers no advantage over 6-MP in maintenance therapy [58,59]. For unknown reasons, 6-MP is more effective when administered in the evening than in the morning. Continuous adaptations of the doses of MTX and 6-MP based on peripheral blood counts are necessary to reduce the risk of relapse, on the one hand, and the risk of infections, on the other [60,61]. There are large interindividual differences in the doses that are tolerated or needed to reduce cell counts. This variability reflects pharmacogenetic differences, for instance in the status of thiopurine methyltransferase, a key enzyme that inactivates thiopurines [60,62]. Allelic differences are associated with reduced activity. Also, large intraindividual differences in doses occur (eg, because of concurrent viral infections). Recently, the major ALL study groups reached consensus on how to adjust the doses of 6-MP and MTX during maintenance so that the white blood cell count remains between 1.5 and 3.0×10^9/L. Routine measurements of liver function are not necessary in patients who do not have symptoms of liver dysfunction.

A meta-analysis of 42 trials showed that both longer maintenance (3 years versus 2 years) and the use of pulses of vincristine and a glucocorticoid during maintenance result in lower relapse rates but increased death rates [63]. The most important factor that has helped reduce relapses and improve survival is the use of an intensive reinduction course at the start of maintenance therapy. Several randomized studies proved the value of reinduction therapy for childhood ALL [64,65]. Attempts to omit reinduction led to a significant increase in relapse rate [66]. More than 50% of patients who were treated without reinduction did not relapse, however, illustrating that not all patients really need this intensification element. The question, of course, is how to identify these patients early on. When an intensive reinduction course is given, neither longer maintenance nor the use of vincristine/glucocorticoid pulses may contribute significantly to a better OS [63].

The results of the meta-analysis do not exclude the possibility that subgroups of patients may benefit from a longer duration of maintenance. Several study groups use longer maintenance therapy for boys than for girls. Reduction of the duration of maintenance below 2 years in a Japanese study led to an increased risk of relapse [67]. This study, however, also demonstrate that not all patients need 2 years of maintenance therapy. Again, the important question is how to identify these patients. It might be that a long maintenance therapy is less effective in high-risk leukemias with a very aggressive behavior, such as *MLL* gene–rearranged ALL, *bcr-abl*–positive ALL, and T-ALL, in which relapses occur relatively early; the more smoldering types of ALL, such as hyperdiploid and *TEL/AML1*-gene rearranged ALL, might benefit more from maintenance therapy.

A recent large, randomized study did not show a benefit for the use of pulses with vincristine and a glucocorticoid in a selected group of patients treated on a Berlin/Frankfurt/Münster regimen [68]. The benefit of these pulses therefore may be found only in studies that use no or a less intensive

reinduction course, such as in the Dutch Childhood Leukemia Study Group-6 study [69] or in studies in which the upfront therapy is relatively mild.

Who should (not) be transplanted?

Autologous SCT is not effective in childhood ALL and therefore should not be performed. A collaborative study of several large study groups has shown that *BCR/ABL*-positive ALL benefits from allogeneic SCT from a matched related donor both in terms of EFS and OS [12]. For other types of donor this benefit was not proven. A comparable analysis for children who had t(4;11) could not detect a beneficial effect of SCT from any type of donor [26]. Recently, a comparison was performed between children who had very high-risk ALL in first remission who were assigned by the availability of a compatible related donor to receive SCT or to receive chemotherapy when no donor was available [70]. ''Very high risk'' was defined in this study by the presence of one or more of the following criteria: failure to achieve complete remission after 5 weeks' therapy, t(9;22) or t(4;11) positivity, a poor prednisone response associated with T-cell phenotype, or a white blood cell count higher than 100×10^e9/L. The 5-year disease-free survival ratewas better for the patients who received SCT from a matched related donor than for those who received chemotherapy. Only one in six of these high-risk patients had a suitable family donor, however. SCT from alternative donors resulted in an inferior outcome. Therefore the role of allogeneic SCT in first complete remission is limited in these very high-risk patients. Another recent study failed to prove a benefit for allogeneic SCT in very high-risk cases [71], whereas the Berlin/Frankfurt/Münster study group showed that high-risk T-cell ALL cases may benefit from SCT [72].

Treatment of adolescents

Four recent reports from four different countries show that outcome for adolescents who have ALL is better when these patients are treated on a pediatric rather than an adult protocol [73–76]. The 5-year EFS of patients aged 15 to 21 years was approximately 30% higher when they were treated according to a pediatric protocol (Table 3). This result could not be explained by differences in immunophenotype and genetic abnormalities, but there seemed to be large differences in the dose intensity used during treatment. The pediatric protocols contained more glucocorticoids, vincristine, L-asparaginase, MTX, and 6-MP. In addition, it is conceivable that the longer delays between different parts of treatment noted in adolescents treated according to the adult protocols might have played a role. It is possible that hematologists have a different approach in managing toxicities because they generally treat older patients who do not tolerate intensive therapy well. Also, the toxicity caused by SCT usually is accepted as part of therapy, whereas adult hematologists have less experience with glucocorticoid- and asparaginase-induced toxicities. In the Dutch study, use of the

Table 3
Outcome of adolescents treated on a pediatric or adult acute lymphoblastic leukemia protocol

Study group [reference]	Patient number	Age category (in years)	5-year event-free survival (%)
United Sates: pediatric [24]	196	16–21	64
United States: adult [24]	103	16–21	38
Dutch: pediatric [23]	47	15–18	69
Dutch: adult [23]	44	15–18	34
French: pediatric [12]	77	15–20	67
French: adult [12]	100	15–20	41
United Kingdom: pediatric [72]	61	15–17	65
United Kingdom: adult [72]	67	15–17	49

adult ALL treatment protocol resulted in both a higher relapse rate and in a higher toxic death rate for adolescents [74].

Side effects

Nearly all chemotherapy side effects seen in children treated for ALL are temporary. The single most important cause of toxic death is infections: 0.5% to 1.5% of patients die from infections during induction therapy, and between 1% and 3% die from infections while in complete remission [77]. Many toxicities result from using a combination of drugs; some, however, are drug specific. Drug-specific toxicities include neuropathy and constipation caused by vincristine, mucositis caused by MTX, diabetes, behavior disturbances, Cushingoid appearance, osteoporosis, and avascular necrosis of bone caused by glucocorticoids, and allergic reactions and thrombosis caused by asparaginase [78].

Toxicity increases with patient age. For example, children older than 10 years have a higher incidence of side effects to glucocorticoids such as avascular necrosis of bone and hyperglycemia, and pancreatitis and thromboembolic complications caused by L-asparaginase [55]. About 5% to 15% of children older than 10 years of age and adolescents experience one or more of these side effects. It has been shown that short pulses of glucocorticoids (5 days) lead to fewer side effects than more continuous schedules with the same cumulative doses of glucocorticoids.

Perspectives

New genomic techniques

The recent sequencing of the human genome and technical advances in high through-put analysis of DNA copy number and mRNA expression now allow a ''molecular portrait'' of leukemia. Gene-expression profiling can be helpful in classifying ALL patients, in revealing new insights into the pathways involved in different genetic subtypes of ALL, and

in identifying new pathways involved in therapy resistance and new therapeutic targets [79].

The first studies using gene-expression profiling showed that known morphologic, immunophenotypic, and genetic subclasses of ALL had specific gene-expression profiles [28,80,81]. Gene-expression profiling may be even more suitable for classifying ALL cases because it takes into consideration the biologic state and genetic progression [82]. Gene-expression patterns have been revealed that are related to in vitro resistance to several classes of individual agents, to clinical outcome, and to cross-resistance to multiple antileukemic drugs [83,84]. These studies, for example, have shown that *MCL-1* overexpression is involved in glucocorticoid resistance in ALL. Modulation of *MCL-1* expression sensitizes ALL cells to glucocorticoids [85].

Bhojwani and colleagues [86] revealed that gene-expression profiles of early relapsed ALL samples were characterized by the overexpression of genes involved in cell-cycle regulation; this finding might identify attractive new targets for therapy. Armstrong [87] and Stam [88] showed high levels of wild-type *FLT3* in *MLL*-rearranged ALL. High levels of *FLT3* are related to a poor outcome [89], and inhibition of this tyrosine kinase is very effective in *MLL*-rearranged ALL cells in vitro [88] as well as in an in vivo mouse model [87]. This finding has led to the design of two different phase I/II studies of these inhibitors in *MLL*-rearranged ALL.

Genome-wide techniques to screen for mutations and amplifications and for single-nucleotide polymorphisms (SNPs) recently have revealed many recurrent genetic alterations that are important for the development of ALL [90–93] and for the sensitivity to chemotherapy. For example, polymorphisms in folate-related genes are related to the MTX sensitivity of ALL cells [94]. Mullighan and colleagues [90] used SNP arrays to reveal that childhood ALL samples show recurrent gene deletions and amplifications including somatic *PAX5* deletion, which is present in about one third of all ALL cases [90]. Overall deletions were more common than amplification, specifically deletions of genes involved in B-cell differentiation, indicating that arrested development is a key feature of leukemia transformation. In the forthcoming years, large-scale studies will analyze the profile of microRNAs in ALL subtypes [95] and the role of newly discovered genetic subtype-specific microRNAs in ALL.

Targeted therapies

Several new targeted therapies may contribute to a further improvement in treatment results in childhood and adolescent ALL (Table 4). The ultimate target of therapy is the leukemogenic fusion product. The best example is the *BCR/ABL* fusion product leading to an abnormal *ABL* tyrosine kinase activity. Imatinib is an effective inhibitor of this kinase [96], but resistance rapidly occurs when it is used as a single agent, mainly because of the selection or development of leukemic subclones with *BCR-ABL* point

Table 4
New targeted therapies for childhood and adolescent acute lymphoblastic leukemia

Drug	Target	Type of ALL
Imatinib	*ABL* tyrosine kinase	*BCR-ABL* fusion, *NUP214-ABL1* fusion
Dasatinib, nilotinib	*ABL* tyrosine kinase (also many mutations), *SRC* kinases	*BCR-ABL* fusion
PKC412, CEP701, other *FLT3* inhibitors	Mutated *FLT3*, wild type over-expressed *FLT3*	*MLL* gene–rearranged ALL, hyperdiploid ALL
Demethylating agents	Hypermethylation	*MLL* gene–rearranged ALL, other subtypes?
Rituximab	CD20	CD20 + (B-lineage) ALL
Epratuzumab	CD22	CD22 + (B-lineage) ALL
Gemtuzumab ozogamicin	CD33	CD33 + ALL
Alemtuzumab	CD52	CD52 + ALL
Forodesine	PNP (purine nucleoside phosphorylase)	T-ALL
Nelarabine		T-ALL

mutations. It therefore seems that imatinib must be combined with standard antileukemic agents to treat *BCR-ABL*–positive ALL effectively. A European randomized study currently is attempting to assess the efficacy and toxicity of the addition of imatinib to all chemotherapy blocks. Resistance to imatinib is caused mainly by the outgrowth of subclones with mutations in the kinase domain of *BCR-ABL* that interfere with imatinib binding. For most mutations, this resistance can be overcome with dasatinib [97] or nilotinib [98]. A pediatric phase I-II study with dasatinib is underway. The very rare subset of T-ALL with *NUP214-ABL1* fusion also may be a suitable group for targeted therapies using these compounds.

The recent finding that half of T-ALL cases have activating mutations of the *NOTCH1* gene provides a rationale for targeted therapies of the NOTCH pathway. Cleavage of the trans-membrane receptor *NOTCH1* by gamma secretase leads to release of the intracellular domain of *NOTCH1* (ICN1), followed by translocation to the nucleus and transcription activation. Inhibitors of ICN1 production and activity seemed to be toxic for T-ALL cells in vitro and have led to a clinical trial of a gamma secretase inhibitor in patients who had refractory T-ALL; however, this trial was stopped because of gastrointestinal side effects. Targeting the enzyme purine nucleoside phosphorylase in T-ALL, especially by forodesine [99], is another strategy that will be tested in childhood ALL in the forthcoming years. Nelarabine is a nucleoside analogue that is converted intracellularly to cytarabine with promising activity as single agent in T-ALL [100,101].

Overexpression of wild-type *FLT3*, especially in *MLL*-rearranged ALL and hyperdiploid ALL, also provides an opportunity for targeted therapies with *FLT3* inhibitors. Another opportunity may be found in the hypermethylation state of *MLL*-rearranged ALL, where the tumor-suppressor gene *FHIT* is silenced by hypermethylation. Re-expression leads to the

killing of infant *MLL*-rearranged ALL cells, and demethylation agents have the same effect [102].

Finally, different monoclonal antibodies, directed against different antigens (CD20, CD22, and CD52), with or without conjugated toxins, are in early clinical studies in childhood ALL.

Host pharmacogenetics

There is no doubt that host polymorphisms in drug-metabolizing genes alter drug levels and target engagement. The ultimate goal of host pharmacogenetic studies is to optimize drug dosing for each patient to achieve maximum treatment efficacy with a minimum toxicity. Germline SNPs determine the toxicity of different antileukemic drugs [103]. The most extensively studied is the gene encoding for thiopurine methyltransferase (*TPMT*) involved in the metabolism of 6-MP. Genetic polymorphisms in *TPMT* correlate with enzyme activity and with both 6-MP toxicity and outcome in ALL. Many other genes are subject to genetic polymorphisms, and the development of tools such as SNP arrays facilitates the studies of many of these polymorphisms simultaneously.

Summary

More than 80% of children who have ALL are cured with contemporary intensive chemotherapy protocols. In the forthcoming decades it will be of great importance to tailor therapy for individual patients according to early response to therapy (mainly by detecting MRD) so that the intensity of therapy can be reduced or augmented. Also, more specific therapy schedules will be developed for immunophenotypic and genetic subclasses of ALL, because it now is apparent that ALL is not a single disease entity but in fact includes different diseases with differing underlying biology and clinical courses. New genomic techniques will lead to the discovery of new molecular genetic abnormalities that will provide more insights into the biology of the different ALL subtypes. New targeted therapy approaches will be developed, and it will be important to investigate how new agents can be incorporated in existing regimens.

References

[1] Greaves M. Infection, immune responses and the aetiology of childhood leukaemia. Nat Rev Cancer 2006;6(3):193–203.

[2] Fischer S, Mann G, Konrad M, et al. Screening for leukemia- and clone-specific markers at birth in children with T cell precursor ALL suggests a predominantly postnatal origin. Blood 2007;110:3036–8.

[3] Conter V, Arico M, Valsecchi MG, et al. Long-term results of the Italian Association of Pediatric Hematology and Oncology (AIEOP) acute lymphoblastic leukemia studies, 1982–1995. Leukemia 2000;14(12):2196–204.

[4] Schrappe M, Reiter A, Zimmerman M, et al. Long-term results of four consecutive trials in childhood ALL performed by the ALL-BFM study group from 1981 to 1995. Berlin-Frankfurt-Munster. Leukemia 2000;14(12):2205–22.
[5] Gaynon PS, Trigg ME, Heerema NA, et al. Children's Cancer Group trials in childhood acute lymphoblastic leukemia: 1983–1995. Leukemia 2000;14(12):2223–33.
[6] Harms DO, Janka-Schaub GE. Co-operative Study Group for Childhood Acute Lymphoblastic Leukemia (COALL): long-term follow-up of trials 82, 85, 89 and 92. Leukemia 2000; 14(12):2234–9.
[7] Kamps WA, Veerman AJ, van Wering ER, et al. Long-term follow-up of Dutch Childhood Leukemia Study Group (DCLSG) protocols for children with acute lymphoblastic leukemia, 1984–1991. Leukemia 2000;14(12):2240–6.
[8] Silverman LB, Declerck L, Gelber RD, et al. Results of Dana-Farber Cancer Institute Consortium protocols for children with newly diagnosed acute lymphoblastic leukemia (1981–1995). Leukemia 2000;14(12):2247–56.
[9] Vilmer E, Suciu S, Ferster A, et al. Long-term results of three randomized trials (58831, 58832, 58881) in childhood acute lymphoblastic leukemia: a CLCG-EORTC report. Children Leukemia Cooperative Group. Leukemia 2000;14(12):2257–66.
[10] Gustafsson G, Schmiegelow K, Forestier E, et al. Improving outcome through two decades in childhood ALL in the Nordic countries: the impact of high-dose methotrexate in the reduction of CNS irradiation. Nordic Society of Pediatric Haematology and Oncology (NOPHO). Leukemia 2000;14(12):2267–75.
[11] Pui CH, Boyett JM, Rivera GK, et al. Long-term results of total therapy studies 11, 12 and 13A for childhood acute lymphoblastic leukemia at St Jude Children's Research Hospital. Leukemia 2000;14(12):2286–94.
[12] Eden OB, Harrison G, Richards S, et al. Long-term follow-up of the United Kingdom Medical Research Council protocols for childhood acute lymphoblastic leukaemia, 1980–1997. Medical Research Council Childhood Leukaemia Working Party. Leukemia 2000;14(12):2307–20.
[13] Tomizawa D, Koh K, Sato T, et al. Outcome of risk-based therapy for infant acute lymphoblastic leukemia with or without an MLL gene rearrangement, with emphasis on late effects: a final report of two consecutive studies, MLL96 and MLL98, of the Japan Infant Leukemia Study Group. Leukemia 2007;21:2258–63.
[14] Pieters R, den Boer ML, Durian M, et al. Relation between age, immunophenotype and in vitro drug resistance in 395 children with acute lymphoblastic leukemia—implications for treatment of infants. Leukemia 1998;12(9):1344–8.
[15] Ramakers-van Woerden NL, Pieters R, Hoelzer D, et al. In vitro drug resistance profile of Philadelphia positive acute lymphoblastic leukemia is heterogeneous and related to age: a report of the Dutch and German Leukemia Study Groups. Med Pediatr Oncol 2002; 38(6):379–86.
[16] Biondi A, Cimino G, Pieters R, et al. Biological and therapeutic aspects of infant leukemia. Blood 2000;96(1):24–33.
[17] Pieters R, Schrappe M, De Lorenzo P, et al. A treatment protocol for infants younger than 1 year with acute lymphoblastic leukaemia (Interfant-99): an observational study and a multicentre randomised trial. Lancet 2007;370(9583):240–50.
[18] Hilden JM, Dinndorf PA, Meerbaum SO, et al. Analysis of prognostic factors of acute lymphoblastic leukemia in infants: report on CCG 1953 from the Children's Oncology Group. Blood 2006;108(2):441–51.
[19] Rots MG, Pieters R, Peters GJ, et al. Role of folylpolyglutamate synthetase and folylpolyglutamate hydrolase in methotrexate accumulation and polyglutamylation in childhood leukemia. Blood 1999;93(5):1677–83.
[20] Heerema NA, Sather HN, Sensel MG, et al. Prognostic impact of trisomies of chromosomes 10, 17, and 5 among children with acute lymphoblastic leukemia and high hyperdiploidy (> 50 chromosomes). J Clin Oncol 2000;18(9):1876–87.

[21] Kaspers GJ, Smets LA, Pieters R, et al. Favorable prognosis of hyperdiploid common acute lymphoblastic leukemia may be explained by sensitivity to antimetabolites and other drugs: results of an in vitro study. Blood 1995;85(3):751–6.
[22] Ramakers-van Woerden NL, Pieters R, Loonen AH, et al. TEL/AML1 gene fusion is related to in vitro drug sensitivity for L-asparaginase in childhood acute lymphoblastic leukemia. Blood 2000;96(3):1094–9.
[23] Stams WA, den Boer ML, Holleman A, et al. Asparagine synthetase expression is linked with L-asparaginase resistance in TEL-AML1-negative but not TEL-AML1-positive pediatric acute lymphoblastic leukemia. Blood 2005;105(11):4223–5.
[24] Stams WA, den Boer ML, Beverloo HB, et al. Sensitivity to L-asparaginase is not associated with expression levels of asparagine synthetase in t(12;21)+ pediatric ALL. Blood 2003;101(7):2743–7.
[25] Frost BM, Froestier E, Gustafsson G, et al. Translocation t(12;21) is related to in vitro cellular drug sensitivity to doxorubicin and etoposide in childhood acute lymphoblastic leukemia. Blood 2004;104(8):2452–7.
[26] Pui CH, Gaynon PS, Boyett JM, et al. Outcome of treatment in childhood acute lymphoblastic leukaemia with rearrangements of the 11q23 chromosomal region. Lancet 2002; 359(9321):1909–15.
[27] Jansen MW, Corral L, van der Velden VH, et al. Immunobiological diversity in infant acute lymphoblastic leukemia is related to the occurrence and type of MLL gene rearrangement. Leukemia 2007;21(4):633–41.
[28] Armstrong SA, Staunton JE, Silverman LB, et al. MLL translocations specify a distinct gene expression profile that distinguishes a unique leukemia. Nat Genet 2002;30(1):41–7.
[29] Dordelmann M, Reiter A, Borkhardt A, et al. Prednisone response is the strongest predictor of treatment outcome in infant acute lymphoblastic leukemia. Blood 1999;94(4): 1209–17.
[30] Ramakers-van Woerden NL, Beverloo HB, Veerman AJ, et al. In vitro drug-resistance profile in infant acute lymphoblastic leukemia in relation to age, MLL rearrangements and immunophenotype. Leukemia 2004;18(3):521–9.
[31] Stam RW, den Boer ML, Meijerink JP, et al. Differential mRNA expression of Ara-C-metabolizing enzymes explains Ara-C sensitivity in MLL gene-rearranged infant acute lymphoblastic leukemia. Blood 2003;101(4):1270–6.
[32] Ramakers-van Woerden NL, Pieters R, Rots MG, et al. Infants with acute lymphoblastic leukemia: no evidence for high methotrexate resistance. Leukemia 2002;16(5):949–51.
[33] Stam RW, van den Heuvel-Eibrink MM, den Boer ML, et al. Multidrug resistance genes in infant acute lymphoblastic leukemia: Ara-C is not a substrate for the breast cancer resistance protein. Leukemia 2004;18(1):78–83.
[34] Thompson PA, Murry DJ, Rosner GL, et al. Methotrexate pharmacokinetics in infants with acute lymphoblastic leukemia. Cancer Chemother Pharmacol 2007;59(6):847–53.
[35] Schrappe M, Arico M, Harbott J, et al. Philadelphia chromosome-positive (Ph+) childhood acute lymphoblastic leukemia: good initial steroid response allows early prediction of a favorable treatment outcome. Blood 1998;92(8):2730–41.
[36] Graux C, Cools J, Michaux L, et al. Cytogenetics and molecular genetics of T-cell acute lymphoblastic leukemia: from thymocyte to lymphoblast. Leukemia 2006;20(9): 1496–510.
[37] Breit S, Stanulla M, Flohr T, et al. Activating NOTCH1 mutations predict favorable early treatment response and long-term outcome in childhood precursor T-cell lymphoblastic leukemia. Blood 2006;108(4):1151–7.
[38] Moorman AV, Richards SM, Robinson HM, et al. Prognosis of children with acute lymphoblastic leukemia (ALL) and intrachromosomal amplification of chromosome 21 (iAMP21). Blood 2007;109(6):2327–30.
[39] Nachman JB, Heerema NA, Sather H, et al. Outcome of treatment in children with hypodiploid acute lymphoblastic leukemia. Blood 2007;110(4):1112–5.

[40] Bassal M, La MK, Whitlock JA, et al. Lymphoblast biology and outcome among children with Down syndrome and ALL treated on CCG-1952. Pediatr Blood Cancer 2005;44(1): 21–8.
[41] Whitlock JA, Sather HN, Gaynon P, et al. Clinical characteristics and outcome of children with Down syndrome and acute lymphoblastic leukemia: a Children's Cancer Group study. Blood 2005;106(13):4043–9.
[42] Tubergen DG, Gilchrist GS, O'Brien RT, et al. Improved outcome with delayed intensification for children with acute lymphoblastic leukemia and intermediate presenting features: a Childrens Cancer Group phase III trial. J Clin Oncol 1993;11:527–37.
[43] Bostrom BC, Sensel MR, Sather HN, et al. Dexamethasone versus prednisone and daily oral versus weekly intravenous mercaptopurine for patients with standard-risk acute lymphoblastic leukemia: a report from the Children's Cancer Group. Blood 2003;101(10): 3809–17.
[44] Mitchell CD, Richards SM, Kinsey SE, et al. Benefit of dexamethasone compared with prednisolone for childhood acute lymphoblastic leukaemia: results of the UK Medical Research Council ALL97 randomized trial. Br J Haematol 2005;129(6):734–45.
[45] Igarashi S, Manabe A, Ohara A, et al. No advantage of dexamethasone over prednisolone for the outcome of standard- and intermediate-risk childhood acute lymphoblastic leukemia in the Tokyo Children's Cancer Study Group L95-14 protocol. J Clin Oncol 2005; 23(27):6489–98.
[46] Kaspers GJ, Veerman AJ, Popp-Snijders C, et al. Comparison of the antileukemic activity in vitro of dexamethasone and prednisolone in childhood acute lymphoblastic leukemia. Med Pediatr Oncol 1996;27(2):114–21.
[47] Ito C, Evans WE, McNinch L, et al. Comparative cytotoxicity of dexamethasone and prednisolone in childhood acute lymphoblastic leukemia. J Clin Oncol 1996;14(8):2370–6.
[48] Duval M, Suciu S, Ferster A, et al. Comparison of Escherichia coli-asparaginase with Erwinia-asparaginase in the treatment of childhood lymphoid malignancies: results of a randomized European Organisation for Research and Treatment of Cancer-Children's Leukemia Group phase 3 trial. Blood 2002;99(8):2734–9.
[49] Moghrabi A, Levy DE, Asselin B, et al. Results of the Dana-Farber Cancer Institute ALL Consortium Protocol 95-01 for children with acute lymphoblastic leukemia. Blood 2007; 109(3):896–904.
[50] Appel IM, Pinheiro JP, den Boer ML, et al. Lack of asparagine depletion in the cerebrospinal fluid after one intravenous dose of PEG-asparaginase: a window study at initial diagnosis of childhood ALL. Leukemia 2003;17(11):2254–6.
[51] Pession A, Valsecchi MG, Masera G, et al. Long-term results of a randomized trial on extended use of high dose L-asparaginase for standard risk childhood acute lymphoblastic leukemia. J Clin Oncol 2005;23(28):7161–7.
[52] Rizzari C, Vasecchi MG, Arico M, et al. Effect of protracted high-dose L-asparaginase given as a second exposure in a Berlin-Frankfurt-Munster-based treatment: results of the randomized 9102 intermediate-risk childhood acute lymphoblastic leukemia study–a report from the Associazione Italiana Ematologia Oncologia Pediatrica. J Clin Oncol 2001;19(5):1297–303.
[53] Amylon MD, Shuster J, Pullen J, et al. Intensive high-dose asparaginase consolidation improves survival for pediatric patients with T cell acute lymphoblastic leukemia and advanced stage lymphoblastic lymphoma: a Pediatric Oncology Group study. Leukemia 1999;13(3):335–42.
[54] Avramis VI, Sencer C, Periclou AP, et al. A randomized comparison of native Escherichia coli asparaginase and polyethylene glycol conjugated asparaginase for treatment of children with newly diagnosed standard-risk acute lymphoblastic leukemia: a Children's Cancer Group study. Blood 2002;99(6):1986–94.
[55] Silverman LB, Gelber RD, Dalton VK, et al. Improved outcome for children with acute lymphoblastic leukemia: results of Dana-Farber Consortium Protocol 91-01. Blood 2001;97(5):1211–8.

[56] Clarke M, Gaynon P, Hann I, et al. CNS-directed therapy for childhood acute lymphoblastic leukemia: childhood ALL Collaborative Group overview of 43 randomized trials. J Clin Oncol 2003;21(9):1798–809.
[57] Matloub Y, Lindemulder S, Gaynon PS, et al. Intrathecal triple therapy decreases central nervous system relapse but fails to improve event-free survival when compared with intrathecal methotrexate: results of the Children's Cancer Group (CCG) 1952 study for standard-risk acute lymphoblastic leukemia, reported by the Children's Oncology Group. Blood 2006;108(4):1165–73.
[58] Harms DO, Gobel U, Spaar HJ, et al. Thioguanine offers no advantage over mercaptopurine in maintenance treatment of childhood ALL: results of the randomized trial COALL-92. Blood 2003;102(8):2736–40.
[59] Vora A, Mitchell CD, Lennard L, et al. Toxicity and efficacy of 6-thioguanine versus 6-mercaptopurine in childhood lymphoblastic leukaemia: a randomised trial. Lancet 2006; 368(9544):1339–48.
[60] Relling MV, Hancock ML, Boyett JM, et al. Prognostic importance of 6-mercaptopurine dose intensity in acute lymphoblastic leukemia. Blood 1999;93(9):2817–23.
[61] Lilleyman JS, Lennard L. Mercaptopurine metabolism and risk of relapse in childhood lymphoblastic leukaemia. Lancet 1994;343(8907):1188–90.
[62] McLeod HL, Relling MV, Liu Q, et al. Polymorphic thiopurine methyltransferase in erythrocytes is indicative of activity in leukemic blasts from children with acute lymphoblastic leukemia. Blood 1995;85(7):1897–902.
[63] Duration and intensity of maintenance chemotherapy in acute lymphoblastic leukaemia: overview of 42 trials involving 12,000 randomised children. Childhood ALL Collaborative Group. Lancet 1996;347(9018):1783–8.
[64] Lange BJ, Bostrom BC, Cherlow JM, et al. Double-delayed intensification improves event-free survival for children with intermediate-risk acute lymphoblastic leukemia: a report from the Children's Cancer Group. Blood 2002;99(3):825–33.
[65] Nachman JB, Sather HN, Sensel MG, et al. Augmented post-induction therapy for children with high-risk acute lymphoblastic leukemia and a slow response to initial therapy. N Engl J Med 1998;338(23):1663–71.
[66] Kamps WA, Bokkerink JP, Hahlen K, et al. Intensive treatment of children with acute lymphoblastic leukemia according to ALL-BFM-6 without cranial radiotherapy: results of Dutch Childhood Leukemia Study Group Protocol ALL-7 (1988–1991). Blood 1999; 94(4):1226–36.
[67] Toyoda Y, Manabe A, Tsuchida M, et al. Six months of maintenance chemotherapy after intensified treatment for acute lymphoblastic leukemia of childhood. J Clin Oncol 2000; 18(7):1508–16.
[68] Conter V, Valsecchi MG, Silvestri D, et al. Pulses of vincristine and dexamethasone in addition to intensive chemotherapy for children with intermediate-risk acute lymphoblastic leukaemia: a multicentre randomised trial. Lancet 2007;369(9556):123–31.
[69] Veerman AJ, Hahlen K, Kamps WA, et al. High cure rate with a moderately intensive treatment regimen in non-high-risk childhood acute lymphoblastic leukemia. Results of protocol ALL VI from the Dutch Childhood Leukemia Study Group. J Clin Oncol 1996; 14(3):911–8.
[70] Balduzzi A, Valsecchi MG, Uderzo C, et al. Chemotherapy versus allogeneic transplantation for very-high-risk childhood acute lymphoblastic leukaemia in first complete remission: comparison by genetic randomisation in an international prospective study. Lancet 2005;366(9486):635–42.
[71] Ribera JM, Ortega JJ, Oriol A, et al. Comparison of intensive chemotherapy, allogeneic, or autologous stem-cell transplantation as postremission treatment for children with very high risk acute lymphoblastic leukemia: PETHEMA ALL-93 Trial. J Clin Oncol 2007;25(1): 16–24.

[72] Schrauder A, Reiter A, Gadner H, et al. Superiority of allogeneic hematopoietic stem-cell transplantation compared with chemotherapy alone in high-risk childhood T-cell acute lymphoblastic leukemia: results from ALL-BFM 90 and 95. J Clin Oncol 2006;24(36): 5742–9.
[73] Boissel N, Auclerc MF, Lheritier V, et al. Should adolescents with acute lymphoblastic leukemia be treated as old children or young adults? Comparison of the French FRALLE-93 and LALA-94 trials. J Clin Oncol 2003;21(5):774–80.
[74] de Bont JM, Holt B, Dekker AW, et al. Significant difference in outcome for adolescents with acute lymphoblastic leukemia treated on pediatric vs adult protocols in the Netherlands. Leukemia 2004;18(12):2032–5.
[75] Deangelo DJ. The treatment of adolescents and young adults with acute lymphoblastic leukemia. Hematology Am Soc Hematol Educ Program 2005;123–30.
[76] Ramanujachar R, Richards S, Hann I, et al. Adolescents with acute lymphoblastic leukaemia: outcome on UK national paediatric (ALL97) and adult (UKALLXII/E2993) trials. Pediatr Blood Cancer 2007;48(3):254–61.
[77] Christensen MS, Heyman M, Mottonen M, et al. Treatment-related death in childhood acute lymphoblastic leukaemia in the Nordic countries: 1992–2001. Br J Haematol 2005; 131(1):50–8.
[78] Caruso V, Iacoviello L, Di Castelnuovo A, et al. Thrombotic complications in childhood acute lymphoblastic leukemia: a meta-analysis of 17 prospective studies comprising 1752 pediatric patients. Blood 2006;108(7):2216–22.
[79] Carroll WL, Bhojwani D, Min DJ, et al. Childhood acute lymphoblastic leukemia in the age of genomics. Pediatr Blood Cancer 2006;46(5):570–8.
[80] Yeoh EJ, Ross ME, Shurtleff SA, et al. Classification, subtype discovery, and prediction of outcome in pediatric acute lymphoblastic leukemia by gene expression profiling. Cancer Cell 2002;1(2):133–43.
[81] Ross ME, Zhou X, Song G, et al. Classification of pediatric acute lymphoblastic leukemia by gene expression profiling. Blood 2003;102(8):2951–9.
[82] Schrappe M. [Medical centers–methods, purpose and benefits]. Z Arztl Fortbild Qualitatssich 2007;101(3):141–5 [in German].
[83] Holleman A, Cheok MH, den Boer ML, et al. Gene-expression patterns in drug-resistant acute lymphoblastic leukemia cells and response to treatment. N Engl J Med 2004; 351(6):533–42.
[84] Lugthart S, Cheok MH, den Boer ML, et al. Identification of genes associated with chemotherapy crossresistance and treatment response in childhood acute lymphoblastic leukemia. Cancer Cell 2005;7(4):375–86.
[85] Wei G, Twomey D, Lamb J, et al. Gene expression-based chemical genomics identifies rapamycin as a modulator of MCL1 and glucocorticoid resistance. Cancer Cell 2006;10(4): 331–42.
[86] Bhojwani D, Kang H, Moskowitz NP, et al. Biologic pathways associated with relapse in childhood acute lymphoblastic leukemia: a Children's Oncology Group study. Blood 2006;108(2):711–7.
[87] Armstrong SA, Kung AL, Mabon ME, et al. Inhibition of FLT3 in MLL. Validation of a therapeutic target identified by gene expression based classification. Cancer Cell 2003; 3(2):173–83.
[88] Stam RW, den Boer ML, Schneider P, et al. Targeting FLT3 in primary MLL-gene-rearranged infant acute lymphoblastic leukemia. Blood 2005;106(7):2484–90.
[89] Stam RW, Schneider P, de Lorenzo P, et al. Prognostic significance of high-level FLT3 expression in MLL-rearranged infant acute lymphoblastic leukemia. Blood 2007;110(7): 2774–5.
[90] Mullighan CG, Goorha S, Radtke I, et al. Genome-wide analysis of genetic alterations in acute lymphoblastic leukaemia. Nature 2007;446(7137):758–64.

[91] Lahortiga I, De Keersmaecker K, Van Vlierberghe P, et al. Duplication of the MYB oncogene in T cell acute lymphoblastic leukemia. Nat Genet 2007;39(5):593–5.
[92] van Vlierberghe P, Meijerink JP, Lee C, et al. A new recurrent 9q34 duplication in pediatric T-cell acute lymphoblastic leukemia. Leukemia 2006;20(7):1245–53.
[93] Van Vlierberghe P, van Grotel M, Beverloo HB, et al. The cryptic chromosomal deletion del(11)(p12p13) as a new activation mechanism of LMO2 in pediatric T-cell acute lymphoblastic leukemia. Blood 2006;108(10):3520–9.
[94] Cheok MH, Evans WE. Acute lymphoblastic leukaemia: a model for the pharmacogenomics of cancer therapy. Nat Rev Cancer 2006;6(2):117–29.
[95] Lu J, Getz G, Miska EA, et al. MicroRNA expression profiles classify human cancers. Nature 2005;435(7043):834–8.
[96] Champagne MA, Capdeville R, Krailo M, et al. Imatinib mesylate (STI571) for treatment of children with Philadelphia chromosome-positive leukemia: results from a Children's Oncology Group phase 1 study. Blood 2004;104(9):2655–60.
[97] Talpaz M, Shah NP, Kantarjian H, et al. Dasatinib in imatinib-resistant Philadelphia chromosome-positive leukemias. N Engl J Med 2006;354(24):2531–41.
[98] Kantarjian H, Giles F, Wunderle L, et al. Nilotinib in imatinib-resistan CML and Philadelphia chromosome-positive ALL. N Engl J Med 2006;354(24):2542–51.
[99] Gandhi V, Kilpatrick JM, Plunkett W, et al. A proof-of-principle pharmacokinetic, pharmacodynamic, and clinical study with purine nucleoside phosphorylase inhibitor immucillin-H (BCX-1777, forodesine). Blood 2005;106(13):4253–60.
[100] Kurtzberg J, Ernst TJ, Keating MJ, et al. Phase I study of 506U78 administered on a consecutive 5-day schedule in children and adults with refractory hematologic malignancies. J Clin Oncol 2005;23(15):3396–403.
[101] Berg SL, Blaney SM, Devidas M, et al. Phase II study of nelarabine (compound 506U78) in children and young adults with refractory T-cell malignancies: a report from the Children's Oncology Group. J Clin Oncol 2005;23(15):3376–82.
[102] Stam RW, den Boer ML, Passier MM, et al. Silencing of the tumor suppressor gene FHIT is highly characteristic for MLL gene rearranged infant acute lymphoblastic leukemia. Leukemia 2006;20(2):264–71.
[103] Kishi S, Cheng C, French D, et al. Ancestry and pharmacogenetics of antileukemic drug toxicity. Blood 2007;109(10):4151–7.