REVIEW ARTICLE

Acute Lymphoblastic Leukemia: Recent Advances in Biology and Therapy

By Richard Champlin and Robert Peter Gale

ACUTE LYMPHOBLASTIC leukemia (ALL) is characterized by excessive accumulation of lymphoblasts and their progenitors. ALL is the most frequent childhood cancer and accounts for approximately 20% of adult acute leukemias. The disease is heterogeneous; the malignant cells express diverse phenotypes and respond variably to chemotherapy. Identification of prognostic factors has allowed classification of the disease into low-, average-, and high-risk forms that require different therapeutic approaches. Substantial advances have been made in treatment. Most children and up to one third of adults are now cured. Several recent reports have addressed important areas of ongoing research. This review summarizes recent advances in classification, biology, prognostic factors, and treatment of ALL.

CLASSIFICATION

ALL can be classified by morphologic and immunologic criteria. The French-American-British (FAB) classification recognizes three morphologic subtypes: L1, L2, and L3. The L1 subtype consists of small uniform lymphoblasts and is the most common subtype in children. The L2 subtype, characterized by large pleomorphic lymphoblasts, is more common in adults. Leukemia cells in the L3 subtype resemble Burkitt lymphoma; this form typically occurs in children or young adults.

ALL can also be classified by immunologic criteria. The leukemic lymphoblasts typically express antigens corresponding to different stages of B- or T-cell development. Approximately 80% of cases of ALL arise from the B-cell lineage. Most of these cases express the common ALL antigen (CALLA), B-cell differentiation antigens, and have undergone immunoglobulin gene rearrangement; surface membrane immunoglobulin is not present. CALLA has recently been identified as a neutral endopeptidase. CALLA-positive ALL can be further classified based on expression of cytoplasmic immunoglobulin. Approximately 80% of cases lack cytoplasmic immunoglobulin and are designated early pre-B. The remaining 20%, which express cytoplasmic immunoglobulin, are termed pre-B. Less than 5% of ALL cases are categorized as mature B-cell type. These cells express surface antigens of mature B cells including surface membrane immunoglobulin; L3 morphology is typical.

Ten percent to 20% of ALL cases arise from the T-cell lineage. These cases express the E-rosette receptor or other T-cell antigens; CALLA usually is absent. In most instances, one or more of the T-cell receptor genes is rearranged. T-cell ALL can be subdivided into early, intermediate, or mature thymocyte types based upon expression of T-cell differentiation antigens. ALL without B- or T-cell features is termed null ALL. The distribution of immunologic subtypes varies between children and adults; children have a higher proportion of B-cell and CALLA positive ALL, whereas adults are more likely to have T-cell and null ALL.

In some instances, the distinction between T- and B-lineage ALL is unclear and the malignant cells may express surface antigens of both subsets. For example, immunoglobulin gene rearrangement occurs in about 10% of cases of T-cell ALL. Conversely, some cases with B-lineage ALL have T-cell receptor gene rearrangement. Possibly, these leukemias reflect transformation of an uncommitted lymphoid progenitor with the potential to differentiate into both T- or B-cells. Overlap between lymphoid and myeloid cells has also been reported. Leukemia cells in 20% of children, and one third of adults with ALL express both lymphoid and myeloid antigens. These cases have been termed hybrid leukemias and may be biphenotypic or bilineal. These leukemias probably develop from a pluripotent progenitor cell capable of both myeloid and lymphoid differentiation or result from aberrant gene expression in a more mature lymphoid or myeloid stem cell.

CYTOGENETIC AND ONCOGENE ABNORMALITIES

Cytogenetic abnormalities are common in ALL. The most frequent translocations in B-lineage ALL are t(1;19),

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t(4;11), t(11;14), and t(9;22). ALL with the L3 phenotype (mature B-cell leukemia) typically exhibits abnormalities similar to Burkitt lymphoma, including t(8;14), t(2;8), or t(8;22) with breakpoints near the immunoglobulin heavy chain locus on chromosome 14 or light chain loci on chromosomes 2 or 22. In T-cell ALL, translocations frequently occur near the site of the α chain gene of the T-cell receptor on chromosome 14, including inv 14, t(14;14), and t(10;14). Inv 14 (q11;q32) can result in juxtaposition of the gene for the T-α receptor joining segment with the immunoglobulin heavy chain locus. In other patients with T-cell malignancies, the translocation involves chromosome 7 at the site of the T-cell receptor beta chain.6q-, 9p- also occur in T-cell ALL. In addition, t(4;11) is common in hybrid leukemias with phenotypic features of monocytes and the B-lymphoid lineage.

The effects of chromosomal rearrangements are intimately linked to rearrangement and altered regulation of cellular oncogenes. These genes, first identified in the context of acutely transforming retroviruses, are thought to play a role in normal cell growth and development by regulating proliferation and differentiation. Abnormal expression of these or closely related genes results in unregulated growth or malignant transformation. Mechanisms of oncogene activation are diverse and include viral transduction, insertional mutagenesis, mutation, amplification, and others. The mechanisms whereby oncogenes might cause ALL are unknown and it is likely that multiple pathways exist. Some oncogene products may function as lymphoid cell growth factors or their receptors; others may regulate DNA replication or cell division. Disruption of the balance between dividing, nondividing cells, and terminally differentiated cells by an uncoupling of proliferation and differentiation is likely to be important.

Retroviruses are a common cause of lymphoid leukemias in animals. Most of these retroviruses lack oncogenes and cause leukemia in a small proportion of infected animals after a variable latency period. Most data indicate that these retroviruses induce leukemia by activating cellular oncogenes. This occurs when the retrovirus inserts into cellular DNA at a specific site in proximity to an oncogene and is referred to as insertional mutagenesis. This process is thought to result in dysregulation of the oncogene.

Acute transforming retroviruses that carry transduced cellular oncogenes can also cause lymphoid leukemias under experimental conditions. Leukemia occurs in a high proportion of infected animals after a brief period. Since these retroviruses are replication defective, they do not cause spontaneously occurring leukemias. A third class of retroviruses that cause lymphoid leukemias include the human T-lymphotrophic viruses (HTLV)-I and II and bovine leukemia virus. These viruses do not insert proximal to cellular oncogenes nor do they carry transduced oncogenes. Their mechanisms of leukemogenesis appear to involve transactivation of normal cellular genes (possibly oncogenes). HTLV-I causes adult T-cell leukemia/lymphoma and HTLV-II is associated with rare cases of T-cell hairy cell leukemia. Retroviruses have not been etiologically linked to the development of acute lymphoblastic leukemia in humans.

Two forms of ALL associated with consistent chromosome abnormalities and altered expression of oncogenes have been studied in detail: ALL associated with t(9;22) and t(8;14). About 5% of children and up to 25% of adults with ALL have the Philadelphia chromosome, produced as a result of the t(9;22) (q34;q11) translocation. This translocation results in a break within the breakpoint cluster region (BCR) gene on chromosome 22 and transfer of some of all of the ABL proto-oncogene from chromosome 9 to a position adjacent to the truncated BCR gene, forming a chimeric BCR-ABL gene. In about half the cases, the rearrangement is similar to that observed in chronic myelogenous leukemia and results in the expression of a 210-kD chimeric BCR-ABL protein. The other half of Philadelphia positive ALL patients have a translocation of ABL to a more 5' region of the BCR gene, which results in expression of a 190 kD BCR-ABL protein. The 190- and 210-kD BCR-ABL gene products have increased tyrosine kinase activity compared to the normal ABL gene product.

It is uncertain whether patients with Philadelphia chromosome-positive ALL and the 210-kD BCR-ABL protein have lymphoid acute phase of chronic myelogenous leukemia (CML) or a distinct disorder. The former notion is supported by the observation that myeloid stem cells in some of these patients also express the BCR rearrangement, and some patients subsequently develop clinical features of the chronic phase of that disease. In other cases, BCR rearrangement appears to be restricted to lymphoid cells.

Mature B-cell ALL and Burkitt lymphoma typically exhibit the t(8;14) (q24;q32), t(2;8) (p11;q24), or t(8;22) (q24;q11) translocation. In these cases, regulation of the MYC oncogene is abnormal. The mechanism for this dysregulation is controversial. It may occur when breaks at different sites within or near MYC place portions of the gene under the influence of regulatory elements of the immunoglobulin heavy or light chain loci or as a result of somatic mutation involving the MYC gene. The recombination between MYC and immunoglobulin genes has been postulated to result from misrecognition of MYC sequences by a recombinase involved in immunoglobulin gene rearrangement.

Other diverse oncogene abnormalities are reported in ALL including mutations, other translocations, and amplification. Mutations in the NRAS gene are reported in about 10% of cases of ALL. Translocations involving oncogenes associated with ALL are shown in Table 1. Examples include ABL in t(9;22), MYC in mature B-cell ALL with t(8;14), and t(8;22), and also in T-cell ALL with t(8;14). BCL2 in B-cell ALL with t(14;18), and TCL-1 in T-cell ALL with t(11;14). These translocations typically juxtapose all or part of the relevant oncogene with a functionally active gene. As a consequence, expression of the oncogene is altered. In some instances, such as Philadelphia chromosome positive ALL, the translocation leads to formation of a novel chimeric gene and fusion protein. In other instances, like BCL2 with the t(14;18) translocation, a
chimeric gene and transcript are formed but the normal BCL2 gene product is expressed in increased levels. Other translocations remain to be investigated. Gene amplification is another mechanism of oncogene activation. This seems to be uncommon in ALL except for a single instance of MYB amplification.

Altered levels of oncogene transcripts are also reported in ALL. MYC and MYB mRNA levels are frequently increased. Abnormalities in FOS, FES, ABL, HRAS, KRAS, FMS, SIS, and SRC mRNA are also reported. Whether these changes are lymphoid-specific or associated with abnormal cell proliferation or differentiation is uncertain. There are few studies of oncogene-related proteins in ALL. P53 expression is increased in ALL cells, but this is unlikely to be leukemia-specific. As indicated, an abnormal 190- or 210-kD ABL-related protein is typical of Philadelphia positive ALL. An 83-kD MYB related protein is reported in a case of ALL. With the exception of Philadelphia positive ALL, it is not known if any of these findings are leukemia-specific.

Although oncogene abnormalities are associated with ALL in humans, there is no direct evidence that they play an etiologic or pathophysiologic role. Nevertheless, activation of similar or identical genes by retroviruses or under experimental conditions can transform hematopoietic cells in vitro and can induce leukemia in animals. If the precise role and mechanism of oncogenes in leukemogenesis can be defined, innovative diagnostic and therapeutic strategies may be forthcoming.

**Lymphoid Stem Cells and Growth Factors**

Lymphocytic leukemias result from abnormal proliferation of lymphocytes and their progenitors. Data in mice and humans indicate that lymphoid and myeloid stem cells arise from a common progenitor. Likewise, T- and B-cells share a common progenitor cell. Most data suggest that B-cell development is more closely linked to myelopoiesis than that of T cells. For example, the Philadelphia chromosome is usually detected in B but not T lymphocytes from persons with CML.

Assays of normal and leukemic lymphoid stem cells in humans have been developed over the past few years. These techniques include growth in semisolid matrices, limiting dilution analyses, plaque-forming assays, and others. The relevance of these assays must be defined by additional studies. These assays are potentially useful in understanding mechanisms of leukemogenesis, evaluation of new drugs, and detecting residual leukemia.

In most instances, in vitro growth of normal and malignant lymphoid cells requires carefully controlled culture conditions. These observations have led to the identification of a family of growth factors that stimulate proliferation and differentiation of B and T lymphocytes. Examples include interleukin-1 (IL-1), interleukin-2 (IL-2), interleukin-4 (IL-4), interleukin-5 (IL-5), and interleukin-6 (IL-6). Negative regulators of lymphopoiesis have also been identified such as transforming growth factor-β. Interferons (IFN) and tumor necrosis factor (TNF) have diverse effects and may stimulate or inhibit lymphoid proliferation under some conditions. The role of any of these factors in the pathogenesis of ALL is uncertain.

**PROGNOSIS**

Prognosis of patients with ALL depends on several interrelated factors including age, initial leukocyte level, cytogenetic abnormality, immune phenotype, CNS involvement, sex, race, extent of lymphadenopathy, and response to therapy. Since most patients with ALL achieve remission, these prognostic factors relate primarily to the duration of remission. It is important to recognize that the impact of prognostic factors depends on the efficacy of therapy; more effective regimens decrease the importance of prognostic variables.

Age is an important but complex risk factor in ALL. Children aged 2 to 10 years have the best prognosis. Poorer results occur in adults and infants younger than 12 months of age. The poor prognosis in infants is most likely related to a higher incidence of undifferentiated and hybrid leukemias. In adults, increasing age is associated with lower remission rates and shorter remissions. When analyzing results of any therapeutic trial, it is important to consider the age limits and distribution since these factors have a major effect on outcome.

Prognosis is inversely related to leukocyte level at diagnosis. Although this is likely a continuous variable, leukocytes greater than 20 or 50 x 10^9/L is often used as a high-risk criterion. Elevated serum levels of lactate dehydrogenase-
nase, IL-2 receptor, and CD8 antigen correlate with higher leukocyte levels and have also been associated with poor prognosis; these data require confirmation.

Cytogenetic abnormalities provide independent prognostic information. In children, hyperdiploidy (>50 chromosomes) is associated with a favorable outcome if translocations are not present. In contrast, pseudodiploidy predicts a poor result. Some specific translocations are associated with a particularly poor prognosis, including t(9;22), t(8;14), and t(4;11).

Immunologic phenotype of the leukemia cells is also of prognostic importance, although recent data suggest its contribution is relatively minor. Immune phenotype differs between children and adults. In children, the CALLA positive subgroup has a relatively good prognosis. The early pre-B (cytoplasmic Ig negative) group is reported to have a better prognosis than the pre-B subset. Mature B-cell ALL has a poor prognosis, but improved results have been reported in recent studies employing therapy similar to that used for Burkitt lymphoma. T-cell ALL is associated with a poor prognosis in children but more favorable results in adults receiving intensive chemotherapy. However, immune phenotype (CALLA v T-cell type) is not an independent variable in many studies; its relationship is largely accounted for by other factors, particularly age and leukocyte count. Several recent studies report improved results in T-cell ALL with intensive therapy underscoring the concept that the importance of prognostic factors diminishes with more effective therapy. Children and adults with a diagnosis of ALL whose leukemia cells express myeloid antigens have a poorer prognosis than those with exclusively lymphoid phenotypes.

Morphologic subtype is of limited prognostic import. In children, the L1 subtype is associated with a better prognosis than L2 in most studies, but this distinction does not have prognostic significance in adults.

Sex is another prognostic factor in ALL. Men have a somewhat poorer prognosis than women which is only partly explained by the risk of testicular relapse. CNS leukemia and extensive lymphadenopathy or hepatosplenomegaly are also adverse risk factors; the latter is often referred to as the lymphoma/leukemia syndrome. Race is another risk factor; blacks have a poorer prognosis than whites. The incidence of ALL is lower in blacks, but the disease typically occurs at a younger age and is usually associated with other unfavorable prognostic factors.

Combinations of independent prognostic factors have been used to identify low- and high-risk groups. Criteria typically include age and leukocyte count. Other variables include CNS involvement, lymphoma syndrome or mediastinal mass, immunophenotype, and cytogenetics. Criteria used to define prognostic categories vary among treatment centers, making direct comparison of results difficult. Recently, a uniform definition of high-risk ALL was proposed.

Response to initial therapy is of major prognostic import. The rapidity of cytoreduction and time to remission are important determinants of remission duration and survival. Patients achieving complete remission in less than 4 weeks have longer remissions than patients who require more extensive therapy. In vitro tests proposed to predict response to chemotherapy include corticosteroid receptors, blast cell DNA content, or reduction of tumor cell growth in clonogenic assays following drug exposure. The value of these assays requires confirmation in prospective studies.

TREATMENT

Modern treatment strategies are often based on risk group assignment. The effectiveness of chemotherapy in low- and standard-risk children has led to attempts to reduce treatment-related morbidity while maintaining favorable results. High-risk children and adults require more intensive therapy to achieve remission and prevent leukemia relapse. In this review we discuss treatment of these three categories of patients separately.

Treatment can be divided into two major phases: remission induction and postremission therapy. Induction chemotherapy is designed to achieve remission. This is defined as reduction in the number of leukemia cells below the level of detection in the blood, bone marrow, and extramedullary sites by conventional techniques, and normalization of hematology. Postremission chemotherapy is designed to eradicate clinically undetectable leukemia cells. Consolidation or intensification chemotherapy refers to early treatment with combinations of drugs of comparable intensity to induction chemotherapy, generally administered in repeated courses over several months. Maintenance chemotherapy refers to lower dose treatment given continuously or repeatedly for several years. In recent studies, relatively intensive chemotherapy regimens involving multiple cycles given for up to 2 to 3 years have been used. The traditional terms consolidation, intensification, and maintenance do not accurately describe these regimens. Although we prefer the term postremission or continuation therapy combined with a specific description of the drugs, doses, and schedules, we use these descriptors in this review.

The CNS is an important site of leukemia involvement in ALL. Furthermore, the blood-brain barrier may shelter leukemia cells from systemic chemotherapy. Involvement of the CNS is uncommon at diagnosis, but it is a frequent site of relapse. CNS relapse is often followed by systemic relapse. Treatment of the CNS is important in preventing local and possibly systemic recurrence. This approach is often termed CNS prophylaxis, but is more accurately called treatment of subclinical disease.

Ideally one would like to individually evaluate the role of each component of postremission therapy, such as consolidation or CNS prophylaxis, on the overall results of treatment. This is difficult for several reasons. First, few controlled, randomized trials analyze the impact of individual treatment components on outcome. Studies frequently introduce new induction schedules simultaneously with changes in consolidation, maintenance, or CNS therapy. Second, the importance of some components, such as CNS prophylaxis, may differ for different risk groups. Third, several components of systemic postremission treatment may interact. For example, use of high-dose methotrexate may obviate the need for additional CNS prophylaxis. The complexity of these vari-
ables and use of different prognostic criteria by the major treatment groups makes critical analysis of results of therapy of ALL difficult. It is probably most useful to consider a "consensus" approach for three groups: low- and standard-risk children, high-risk children, and adults. From available data it is not possible to draw firm distinctions in the treatment for low- and standard-risk children, so we consider them jointly as standard-risk ALL in this review.

Standard-Risk Children

Treatment with vincristine and prednisone induces remission in over 90% of children with standard-risk ALL. The addition of L-asparaginase or an anthracyline, such as daunorubicin or doxorubicin, marginally increases remission rate, but remission duration is prolonged.

It is important to define the most effective, least toxic continuation chemotherapy for children with standard-risk ALL. Most centers use a combination of daily 6-mercaptopurine and weekly methotrexate. Male children are usually treated for 3 years and female children for 2 years. Sixty percent to 80% of children with standard-risk ALL remain in continuous remission for 5 or more years with this type of therapy. Many centers also give intermittent pulses of vincristine and prednisone, although there is no convincing data that postremission therapy with these agents is beneficial.

Intensive early or late intensification chemotherapy has not convincingly improved remission duration or the proportion of long-term survivors in standard-risk ALL when compared with conventional maintenance chemotherapy, although results of recent studies involving intermediate- or high-dose methotrexate-based therapy appear encouraging for improvement in disease-free survival.

Treatment of the CNS is important in preventing CNS relapse. Several approaches are used. Most common is the combination of cranial radiation and intrathecal methotrexate, given shortly after achievement of remission. This regimen reduces the rate of relapse in the CNS from 60% to 10%, thereby increasing the proportion of disease-free survivors.

Use of intrathecal methotrexate alone is less effective to prevent CNS relapse, but may produce identical long-term survival. Cranial radiation can cause neurologic defects, learning disabilities, and abnormalities of growth and development in children. Less toxic alternatives to prevent CNS leukemia have been evaluated. Cranial radiation at a dose of 18 Gy is as effective as 24 Gy in standard-risk children but may be inadequate in high-risk children.

Because systemic treatment with intermediate or high doses of intravenous (IV) methotrexate produces therapeutic drug levels in the CSF, there is considerable interest in the use of this agent to prevent CNS relapse. These regimens are effective as CNS prophylaxis in standard-risk children and reduce systemic relapses as well.

There is recent concern that the nonintensive therapies used to treat children with standard-risk ALL may result in a higher incidence of late relapses than observed in high-risk children receiving more intensive post remission treatment. Because of this, some have suggested that standard- and high-risk groups should receive identical intensive postremission therapy; this is a minority viewpoint.

High-Risk Children

Optimal therapy of children with high-risk ALL is controversial, and analysis is complicated by lack of a consistent definition of high-risk disease. Recent trials, beginning with LSA2-L2 protocol and extending to recent trials of the Berlin, Frankfurt, Munster group, the Children’s Cancer Study Group, and the Pediatric Oncology Group use a similar strategy in which intensive therapy is given for several months followed by somewhat less intensive therapy. In some studies, the initial intensive phase is divided into induction and consolidation; in others there is no distinction. Attempts to separate induction and consolidation chemotherapy are sometimes inaccurate because of the difficulty in determining when remission is achieved under these circumstances. These studies share the concept of early use of myelosuppressive drugs in high doses for several months. This contrasts with treatment of standard-risk ALL, where remission can generally be achieved using nonmyelotoxic or only modestly myelotoxic therapy. Recent studies in high-risk ALL have further intensified initial therapy by adding cyclophosphamide, high-dose methotrexate, teniposide, etoposide, or cytarabine. The contribution of these drugs to standard regimens using three or four drugs is unknown. Nevertheless, results using intensive treatment have improved markedly since 1980, and most current protocols use five or six drugs in the initial phase of therapy.

Optimal postremission therapy for high-risk children is also unknown. Most data suggest that intensive chemotherapy is needed to prevent or delay leukemia relapse. Recent studies have generally involved cyclical high-dose intensification for 2 to 3 years with combinations of active drugs such as teniposide, cytarabine, conventional or high-dose methotrexate, cyclophosphamide, 6-thioguanine, 6-mercaptopurine, nitrosoureas, anthracyclines, L-asparaginase, or others. Intermittent pulses of vincristine and prednisone are usually included. Other studies use standard maintenance chemotherapy combined with cycles of intensive chemotherapy at 1- to 2-month intervals. These cycles are often referred to as reinduction if the therapy used is similar to that used for remission induction, or intensification if new drugs are used. No randomized trials have been performed evaluating these alternative approaches. Recent studies involving intensified early postremission therapy in high-risk children report improved disease-free survival ranging from 60% to 80%; these data are comparable to those achieved in standard-risk children.

CNS prophylaxis is also important in treatment of children with high-risk ALL. These children have a higher incidence of CNS relapse than the standard-risk group and require more intensive local therapy. Most data indicate that children with high-risk ALL require 24 Gy cranial radiation plus intrathecal methotrexate or extended intraventricular or intrathecal therapy to prevent CNS and systemic relapses.

In summary, it is difficult to define which component(s) of modern intensive therapy of high-risk children with ALL is responsible for recent improvements in leukemia-free survival. Each protocol has its advocates, and it is difficult to
demonstrate convincing differences. It is also difficult to know which drugs are important in postremission therapy. This uncertainty, combined with the risks of intensive therapy, make randomized controlled trials necessary. In our opinion, effective therapy of children with high-risk ALL is likely to resemble that for other childhood malignancies where early intensive treatment is the most important determinant of outcome.

**ALL in Adults**

Adults with ALL have a poorer prognosis than children with ALL. The overall treatment approach is similar to that used for high-risk children.\(^3\)\(^,\)\(^8\)\(^,\)\(^34\) Vincristine and prednisone produces remissions in approximately 60% of adults.\(^1\)\(^1\)\(^,\)\(^1\)\(^2\) Addition of an anthracycline, usually daunorubicin, increases remission rates from 70% to 80% and is probably more effective than L-asparaginase.\(^1\)\(^3\)\(^,\)\(^1\)\(^4\) Several centers have attempted to further intensify therapy by adding cyclophosphamide or L-asparaginase\(^1\)\(^5\)\(^,\)\(^1\)\(^6\); there is no definitive evidence that this improves results.

The optimal postremission chemotherapy in adults is uncertain. Although best results are reported with intensive consolidation treatment, the efficacy of this approach has not been established in controlled trials. Recent studies indicate 3- to 5-year actuarial leukemia-free survival in 20% to 35% of unselected patients.\(^2\)\(^,\)\(^3\)\(^,\)\(^8\)\(^,\)\(^3\)\(^4\)\(^,\)\(^3\)\(^5\)\(^,\)\(^3\)\(^6\) Analysis of these data are complex and require consideration of prognostic factors. For example, results improve if patients with the Philadelphia chromosome are excluded. Likewise, because of the influence of age, studies involving primarily adolescents have better results than those with older patients. Controlled trials evaluating efficacy of individual components of postremission chemotherapy are also lacking, and it is difficult to determine which are useful. There is no convincing data that any particular combination is most effective. Specifically, there are no definitive data that inclusion of drugs such as high-dose cytarabine, mitoxantrone, teniposide, or etoposide improves results. The optimal duration of postremission therapy is likewise unknown. In a recent study, intensive treatment for 6 months was as effective as 2 to 3 years of therapy.\(^1\)\(^3\) Following completion of intensification, most patients have received standard continuation chemotherapy with relatively low doses of methotrexate and 6-mercaptopurine; the efficacy of this approach is uncertain in adults.

The value of CNS prophylaxis in adults is controversial. The likelihood of meningeal leukemia is considerably lower in adults than in children,\(^8\) although groups at high risk for CNS involvement can be identified.\(^1\)\(^7\) CNS prophylaxis has typically consisted of cranial radiation and intrathecal methotrexate or intrathecal methotrexate alone.\(^1\)\(^8\) Several recent trials in adults using high-dose systemic methotrexate and/or cytarabine without radiation reported low rates of CNS relapse.\(^1\)\(^9\) Intensive intraventricular chemotherapy via an Omaya reservoir is also reported to be effective.\(^1\)\(^5\) In one randomized study, cranial radiation and intrathecal methotrexate decreased the risk of meningeal leukemia but did not improve survival, perhaps because of a poor overall outcome due to systemic relapses.\(^1\)\(0\) Furthermore, persons not receiving CNS prophylaxis who did not relapse within 2 years had no late meningeal relapses. These data suggest that the CNS is a less important leukemia sanctuary in adults than children. We believe that treatment to prevent CNS relapse involving either cranial radiation and intrathecal methotrexate, or systemic high-dose methotrexate and/or cytarabine, should be given to adults with ALL. This is particularly true of patients with a high risk of meningeal relapse, such as patients with initial leukocytes exceeding 50 to 100 \(\times\) \(10^9\)/L.

**Detection of Residual Leukemia**

A major limitation in the therapy of ALL is the inability to detect minimal residual leukemia. This necessitates continued treatment for an arbitrary length of time. Recently, several techniques have been proposed to detect subclinical leukemia, including studies of clonal immunoglobulin or T-cell receptor rearrangements,\(^1\)\(^4\) immunoglobulin idiotype,\(^1\)\(^2\) growth of putative leukemia progenitor cells in clonogenic assays,\(^6\)\(^,\)\(^1\)\(^3\) or assay of leukemia specific gene rearrangements such as BCL2 or BCR-ABL.\(^1\)\(^4\)\(^,\)\(^1\)\(^5\) The value of these approaches is uncertain. Most only marginally increase the sensitivity of detection of leukemia cells. The polymerase chain reaction, which can amplify genomic DNA sequences in vitro, holds considerable potential and may be capable of detecting clonal genetic rearrangements in a small number of residual leukemia cells.\(^1\)\(^4\)\(^,\)\(^1\)\(^5\) Its clinical value has not been tested in ALL.

**TREATMENT OF LEUKEMIA RELAPSE**

Despite modern chemotherapy, many patients with ALL relapse in the bone marrow or other sites. The leukemia cells at relapse typically exhibit similar cytogenic and phenotypic characteristics as at the time of diagnosis.\(^1\)\(6\) Occasionally, the relapse cells exhibit a different phenotype or cytogenetic abnormality, suggesting clonal evolution or development of a second leukemia. Some patients receiving bone marrow transplants have had leukemia recurrence in donor cells.\(^1\)\(7\) For example, in a recent report, B-cell ALL developed in a patient where initial leukemia was T-cell ALL.\(^1\)\(8\) These donor relapses, albeit rare, have important implications for understanding the pathogenesis of ALL.

Relapse can occur in the bone marrow or in extramedullary sites, most commonly the CNS\(^1\)\(9\) or testes.\(^1\)\(5\) Patients with isolated extramedullary relapse are at high risk for subsequent bone marrow relapse and require local treatment followed by systemic reinduction therapy.\(^1\)\(6\) Approximately 20% of children with isolated CNS relapse have achieved long-term leukemia-free survival.\(^1\)\(1\)\(^0\),\(^1\)\(^2\),\(^1\)\(^4\)\(^,\)\(^1\)\(^9\)

The prognosis of patients with bone marrow relapse depends on the duration of first remission.\(^1\)\(5\)\(^,\)\(^1\)\(^4\) Longer initial remissions are associated with a higher probability of achieving a second remission and longer second remissions. Children whose initial remissions exceed 3 years or who relapse more than 6 months after completing continuation therapy have an 80% probability of achieving a second remission. About 40% of these children become long-term leukemia-free survivors when retreated with intensive postremission therapy.\(^1\)\(5\) In contrast, children who relapse within 18 months or while receiving continuation chemotherapy...
have a worse prognosis. Although many achieve second remission, it is generally brief, and fewer than 5% achieve long-term leukemia-free survival. The most important predictive factor appears to be relapse on or off therapy, rather than first remission duration. The prognosis of adults with bone marrow or CNS relapse is poor; only about one half achieve second remissions and median remission duration is less than 6 months.\(^{63}\)

Treatment with drugs used for initial induction chemotherapy is often effective in achieving second remissions, particularly in children. The combination of vincristine, prednisone, an anthracycline, and L-asparaginase may be more effective than three drugs.\(^{152}\) The combination of teniposide and cytarabine induces remissions in approximately one half of patients unresponsive to vincristine- and prednisone-based therapy.\(^{155}\) Etoposide and cytarabine also appear effective.\(^{156}\) Intermediate dose methotrexate combined with L-asparaginase is effective in approximately 30% of cases.\(^{157}\) High-dose methotrexate,\(^{158}\) amsacrine,\(^{159}\) high-dose cytarabine,\(^{160}\) mitoxantrone,\(^{161}\) and idarubicin\(^{162}\) have limited activity as single agents, producing complete remissions in fewer than 20% of patients. Recently, the combination of high-dose cytarabine and amascrine was reported to produce remissions in 27 of 36 patients (75%) with relapsed or refractory ALL.\(^{163}\) Postremission treatment with teniposide and cytarabine,\(^{156}\) or the combination of methotrexate and L-asparaginase\(^{154}\) results in median remission duration of 6 months to 1 year.

These conclusions on treatment of relapse are based on data from children treated with less intensive initial regimens used prior to 1983. This situation may change with modern regimens involving more intensive, prolonged therapy.

**BIOLGIC THERAPIES**

Although initial studies suggested that nonspecific immunotherapy with bacillus Calmette-Guerin (BCG), levamisole, or similar agents prolonged remissions in ALL,\(^{164}\) considerable data in children and adults now indicate that this notion is incorrect. Randomized trials of nonspecific immunotherapy using several approaches indicate no benefit.\(^{80,165}\)

Monoclonal antibodies, (MoAbs) directed to cell surface antigens present on leukemia cells have been evaluated for therapy of advanced ALL. Anti-CALLA or anti-T-cell antibodies result in rapid but transient reductions in circulating lymphoblasts.\(^{166,167}\) Although leukemia cells in the bone marrow react with these antibodies, most are not lysed, and substantial cytoreduction is rare. In some cases there is modulation of the target antigen. MoAbs have been recently conjugated with toxins (immunotoxins) or with radioisotopes\(^{168}\) to improve the therapeutic response. These approaches require critical evaluation.

Alpha interferon (α-IFN) has also been studied as an immunotherapeutic agent; only marginal activity was observed.\(^{169}\) Recently, a randomized study evaluated the use of human leukocyte IFN following bone marrow transplantation (BMT) in patients with ALL. Leukemia relapse was reduced in the IFN group, but survival was not improved;\(^{170}\); this result requires confirmation. Decreased relapses in this setting could be related to effects of IFN on immunologic recovery or augmentation of the graft-ve-leukemia effect associated with allogeneic BMT rather than direct antileukemia activity.

It is possible to envision several uses of lymphoid and hematopoietic growth factors in leukemia therapy. For example, factors that stimulate leukemia cells to proliferate might increase their sensitivity to cell cycle-specific chemotherapy. Factors might also be used to induce differentiation and maturation of lymphoblasts to mature, nondividing cells,\(^{171}\) or to enhance host immunologic antileukemic mechanisms. Clearly these studies are at an early stage. The likelihood of success will depend on understanding the mechanisms of leukemogenesis, identification and molecular cloning of these factors, and therapeutic application of agents with relatively specific effects on leukemia cells.

Recently, a number of recombinant human myeloid hematopoietic growth factors such as granulocyte-macrophage colony-stimulating factor (GM-CSF) and granulocyte colony-stimulating factor (G-CSF) have also been identified and molecularly cloned.\(^{172,173}\) Parenteral treatment with these agents shortens the period of granulocytopenia after myelotoxic chemotherapy and may allow further intensification of treatment.\(^{174,175}\) Studies in animals suggest that IL-3 in combination with GM-CSF or G-CSF further enhances the rate of hematologic recovery.\(^{176}\) Other factors may also be useful, including IL-1 and IL-6, which act synergistically with IL-3, G-CSF, and GM-CSF.\(^{177}\)

**BONE MARROW TRANSPLANTATION**

Radiation and most chemotherapeutic drugs produce dose-dependent antileukemia effects.\(^{178}\) Maximally tolerated doses of these agents are often limited by toxicity to normal bone marrow cells and can be substantially increased when treatment is followed by transplantation of normal bone marrow cells. Usually an HLA-identical sibling is the transplant donor. BMT is more effective than conventional dose chemotherapy in eradicating leukemia in most instances.\(^{179}\) This effect is complex. Some of the enhanced antileukemia activity is likely related to high-dose therapy. There is also evidence of an additional antileukemia effect associated with allogeneic BMT.\(^{180-182}\) This may be separated into three elements: an antileukemia effect of graft-ve-host disease, a separate antileukemia effect of T cells, and possibly a specific graft-ve-leukemia effect.\(^{183}\)

Over 2,000 allogeneic bone marrow transplants have been performed for patients with ALL. Results depend on remission status and patient age.\(^{184-189}\) Results in children are generally superior to those achieved in adults. Patients older than 45 to 50 years of age are not generally considered candidates for BMT.

Transplants for advanced ALL in relapse following high-dose cyclophosphamide and total body radiation (TBI) result in 10% to 20% long-term leukemia-free survival. These results are clearly superior to alternative treatments, but only a small fraction of patients benefit. The major cause of treatment failure is recurrent leukemia with actuarial relapse rates exceeding 60%.

Bone marrow transplants in second remission have substantially better results because of a lower relapse rate; 2- to
4-year disease-free survival is 30% to 40%.

Better results are reported in children than adults. Two recent studies in children with ALL in second remission reported 5-year disease-free survival rates of 40% and 64%. In several studies, prognostic factors at diagnosis and length of initial remission correlated with the risk of relapse following BMT. Data from several studies in children and adolescents receiving bone marrow transplants during second or later remission indicated improved disease-free survival with transplants compared with chemotherapy, 30% vs less than 10%. Recent analyses from the International Bone Marrow Transplant Registry and elsewhere compared results of chemotherapy and bone marrow transplantation in children with ALL in second remission. Transplants were superior to chemotherapy in those who relapsed within 18 months of achieving first remission. Children relapsing after a first remission of 18 to 36 months showed a slight but nonsignificant advantage with BMT, whereas those relapsing more than 36 months after achieving first remission or after completing maintenance chemotherapy had similar results with both therapies. These data suggest that children in second remission who have had long first remissions should probably receive chemotherapy, with bone marrow transplants reserved for those who subsequently relapse. Adults with recurrent ALL have a poor prognosis with chemotherapy and should probably receive BMT in second remission if a suitable donor is available, regardless of the length of their initial remission.

The best results with transplants have been reported in patients in first remission. Typically these are adults or children with high-risk features such as B- or T-cell ALL, CNS involvement, high leukocyte levels, or adverse cytogenetic abnormalities. Relapse rates in this setting are relatively low, 20% to 30%, and survival is approximately 50%. These data raise the important question of which, if any, children or adults should receive bone marrow transplants while in first remission. Children with standard-risk ALL have a relatively good prognosis with chemotherapy and are not candidates. Since recent studies report approximately 70% extended survival with chemotherapy for children with high-risk ALL, it is also reasonable to postpone BMT in this group until after relapse. Children younger than 6 months of age and those with the Ph-chromosome have poor prognosis with chemotherapy and may be transplant candidates. Transplants are technically difficult in infants, however, because of the toxicity of high-dose chemotherapy and radiation.

There is considerable controversy as to whether adults with ALL in first remission should be treated with BMT or postremission chemotherapy. From available data, it is uncertain whether BMT in first remission is superior to initial chemotherapy, with bone marrow transplants reserved for patients who relapse. This question is best addressed in a controlled trial. Some data suggest that adolescents and adults with Philadelphia chromosome-positive ALL may have more favorable results with transplantation.

Recurrent leukemia is a major problem in patients with ALL who receive bone marrow transplants. New conditioning regimens using fractionated TBI with high doses of cytarabine or etoposide, hyperfractionated radiation and cyclophosphamide, or the combination of busulfan and cyclophosphamide report relatively low relapse rates. None of these regimens have been shown to be convincingly superior. Since graft-v-host disease is associated with a lower risk of relapse, its intentional induction was attempted to provide an additional antileukemia effect; this was not successful. Recent data from the International Bone Marrow Transplant Registry suggest that methotrexate prophylaxis for graft-v-host disease with or without corticosteroids is associated with a lower relapse rate than the use of cyclosporine; further study is warranted.

Patients who relapse after BMT are candidates for reinduction chemotherapy; approximately half achieve remission and occasional persons are long-term survivors. Children and patients relapsing more than 1 year after transplantation have the best prognosis. Second bone marrow transplants should be considered in selected patients.

BMT may be associated with serious complications such as toxicity of chemoradiotherapy, graft-v-host disease (GVHD), and infections including interstitial pneumonia. Recent attempts to prevent GVHD by depleting T lymphocytes from the donor bone marrow substantially reduces the incidence and severity of GVHD, but increases the risk of graft rejection and probably of recurrent leukemia. Presumably, donor-derived T-cells are necessary to enhance engraftment and mediate an antileukemia effect. Combinations of immunosuppressive drugs, such as cyclosporine plus methotrexate or prednisone, may also reduce GVHD; their impact on leukemia relapse and survival are uncertain.

Only one third of patients with ALL have an HLA-identical sibling. Recently, some patients have received transplants from HLA-partially matched related donors or from HLA-matched or partially matched unrelated donors. These transplants are associated with a higher risk of graft rejection, GVHD, and infections. Nevertheless, some preliminary encouraging results have been reported.

**Autologous BMT**

There is considerable interest in the use of autologous bone marrow transplants for patients with ALL. Bone marrow is collected and cryopreserved while the patient is in remission. High-dose chemotherapy and/or radiation are administered at a later time, followed by infusion of the cryopreserved bone marrow. This approach is applicable to persons without an HLA-identical donor and avoids the problems of graft rejection and GVHD. The incidence of interstitial pneumonitis is also markedly decreased.

One major concern with autologous marrow transplants for ALL is the high likelihood of residual leukemia cells in the cryopreserved, remission bone marrow. It may be possible to eliminate these cells by ex vivo treatment using physical, immunologic, or pharmacologic techniques. Several studies have used antisera or MoAbs reactive against B- or T-lymphocyte differentiation antigens present on leukemia cells. Antibody-bound leukemia cells were eliminated by complement, by conjugation of the antibodies to toxins such as ricin (immunotoxins), or by use of an iron-bound antibody followed by magnetic depletion.
ACUTE LYMPHOBLASTIC LEUKEMIA

Other approaches include treating the bone marrow with drugs such as mafosfamide or etoposide,\textsuperscript{217,222} or by light exposure following incubation with photoactive dyes.\textsuperscript{223} Combinations of these modalities are reported to further reduce leukemia cell contamination.\textsuperscript{224}

There are important conceptual limitations to the use of these techniques to remove leukemia cells. First, these methods rarely eliminate more than 3 to 4 logs of leukemia cells. Second, leukemia cells are heterogeneous in cell surface antigen expression and drug sensitivity. Although antibodies or drugs may eliminate most leukemia cells, they may not affect leukemia progenitor cells.\textsuperscript{225} The efficacy of depletion is difficult to evaluate in vitro since assays of leukemia progenitor cells are imperfect and have not yet shown to be of biologic import. The clinical value of ex vivo treatment of the autologous bone marrow to prevent relapse following transplantation has not been documented, and will be very difficult to evaluate until more effective preparative regimens are developed to eliminate leukemia in the recipient.

Since allogeneic BMT in patients with advanced ALL is associated with a >60% actuarial relapse rate, it is not surprising that relapse rates following autologous transplants in this setting are 80% to 100%.\textsuperscript{218,226} Leukemia relapse may occur because of residual leukemia cells in the patient, the cryopreserved bone marrow, or both. Given the relatively high relapse rates with allogeneic marrow transplants, the major cause of relapse is likely residual systemic leukemia in the recipient. In addition, the graft-v-leukemia effects of allogeneic BMT are not expected following autologous transplants. Consistent with this concept, recipients of bone marrow transplants from genetically identical twins, a model for autotransplants, seem to have a higher relapse rate than allograft recipients.\textsuperscript{219}

Substantial numbers of patients with ALL in second or later remission have received autologous transplants using antibody-treated remission bone marrow cells.\textsuperscript{187,216,218,226} Disease-free survival is about 20% at 3 years. These results were not significantly different than with allogeneic bone marrow transplants in one study. Relapses were more frequent with autologous transplants but were not higher than in allograft recipients without GVHD.

Autologous bone marrow transplants have been primarily used for ALL patients in second remission. The best results with autologous bone marrow transplants are reported in patients with favorable prognostic factors, such as those who relapse after a long initial remission or with isolated extramedullary relapse; many of these individuals are predicted to respond similarly to chemotherapy.\textsuperscript{191} Results of autologous BMT in patients who relapse early, while receiving postremission chemotherapy, are less encouraging although some long-term survivors are reported.\textsuperscript{191} It is presently uncertain whether results with autologous BMT are superior to those achievable by intensive chemotherapy. Some adults with ALL have received autologous bone marrow transplants while in first remission\textsuperscript{227,228}; results have been variable and there is no convincing evidence that this approach is superior to postremission chemotherapy. Prospective randomized studies are needed.

CONCLUSION

There has been considerable progress in the biology and therapy of ALL. Further advances require development of innovative, effective, less toxic treatments involving chemotherapy, BMT, and biologic therapies. Improvement in detection of minimal residual disease should help direct the intensity of postremission therapy. Recent advances in understanding the pathogenesis of some forms of ALL will hopefully allow development of more selective therapies.

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REFERENCES

26. Mufti GJ, Hamblin TJ, Ossier DG, Johnson S: Common ALL with pre B cell features showing (8;14) and (14;18) chromosome translocations. Blood 62:1142, 1983
31. Dubé ID, Raimondi SC, Pi D, Kalousek DK, Johnson S: Common ALL with pre B cell features showing (8;14) and (14;18) chromosome translocations. Blood 62:1142, 1983
43. Poiesz BJ, Ruscetti FW, Gazdar AF, Bunn PA, Minna JD, Gallo RC: Detection and isolation of type C retrovirus particles from fresh and cultured lymphocytes of a patient with cutaneous T cell lymphoma. Proc Natl Acad Sci USA 77:7415, 1980
51. Clark SS, McLaughlin J, Crist WM, Champlin RE, Witte ON: Unique forms of the abl tyrosine kinase distinguish Ph-positive CML from Ph'-positive ALL. Science 235:87, 1987
56. Taub R, Moulding C, Battey J, Murphy W, Vaiscek T, Lenoir GM, Leder P: Activation and somatic mutation of the...


Eight years' experience with cranial irradiation and intrathecal methotrexate. Blood 61:297, 1983


219. Horowitz MM: Personal communication, March 1989
Acute lymphoblastic leukemia: recent advances in biology and therapy [see comments]

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