The Acute Leukemias

Biologic, Diagnostic, and Therapeutic Determinants

edited by

Sanford A. Stass

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The Acute Leukemias

HEMATOLOGY

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Series Introduction

For most of this century hematology has followed a pattern of major scientific discoveries, improved understanding of disease, and rapid application of new knowledge in the clinic. The rate of advance continues at an accelerating pace, so that all but the most zealous have difficulty in keeping up with the literature in even a limited area of specialized interest. As the explosive development of knowledge continues apace, it is a continuing challenge to keep abreast of significant new developments as they impact on clinical and laboratory hematology. The Marcel Dekker, Inc. Hematology series is designed to help in this respect, by providing up-to-date and expert presentations on important subject areas in our field. It is hoped that these works, both individually and collectively, will become important volumes for updating information and for reference for the clinician, investigator, teacher, and student, and in this manner contribute to the advancement of hematology.

This volume on the acute leukemias is the sixth in this Hematology series. Like its predecessors, it presents the current state of the art of the topic, bringing together the many facets of the subject during a stage of rapid advance in the field. In perspective, we are nearing the centennial of the publication in 1889 by Wilhelm Ebstein, the German physician, which clearly identified acute leukemia as an entity distinct from chronic leukemia. The subsequent slow and tortuous progress in subclassifying the acute leukemias by cell type was punctuated by the discovery in midcentury of chemotherapeutic agents causing remission of the disease by such pioneers as Sidney Farber and George Hitchings and their colleagues. This event, coupled with the advances in cell biology and pathology, provided the stimulus and the methods for better characterization of the cells of acute leukemia. The advent of the new immunology and molecular genetics and

the application of these new technologies to the study of leukemic cells added still new dimensions for study. The results of this new focus and their applications to clinical management of the acute leukemias are expertly covered in this volume by a distinguished group of contributors under the able leadership of the editor, Sanford A. Stass.

Kenneth M. Brinkhous
Series Editor

Preface

The series to which this book belongs is intended to discuss the spectrum of current directions and knowledge in hematology. This particular volume is focused on one of the most dynamic areas of hematology: characterization of acute leukemia and its biologic, diagnostic, and therapeutic determinants.

In the past few years there has been a rapid expansion in our ability to characterize acute leukemia. This has resulted from the endeavors of scientists from multiple disciplines applying their expertise to the study of leukemic cells. Fields such as immunology, cytogenetics, biochemistry, and molecular biology have enabled detailed characterization of acute leukemia and opened new vistas of understanding never before imagined. In the last five years the biologic revolution in the study of acute leukemia has influenced clinical and basic scientists alike. Such knowledge has directly translated into better and more sensitive methods for the diagnosis of acute leukemia and a better understanding of the process of leukemogenesis. Furthermore, it is clear that the refinement of our ability to characterize and diagnose acute leukemia has resulted in the identification of nonrandom phenotypic and biologic features which have important clinical and prognostic implications. Information developed in the laboratory has rapidly become relevant at the bedside. Thus, in order for basic scientists, pathologists, hematopathologists and clinical and laboratory hematologists to keep pace with and understand future developments and applications in acute leukemia, a clear understanding of recent progress in the study of the characterization of acute leukemia is essential.

This book unifies the current complex and multifaceted approach to the characterization of acute leukemia and places in perspective the usefulness of the divergent studies that are currently available for evaluation of the leukemic cell. Subjects discussed by distinguished experts include specialized morphologic and

cytochemical studies, flow cytometry, cytogenetics, immunologic phenotyping, biochemical markers, molecular biology, cytogenetics, and therapeutic implications of leukemic cell characterization. DNA technology, an explosive area of investigation, has provided one of the newest and most precise means for studying and classifying leukemic cells using oncogene and lineage-associated probes. The merging of molecular biology with the studies on nonrandom chromosomal findings, which are important for the classification and prognosis of acute leukemia, has greatly benefited our understanding of the leukemic cell. The insights from such investigations have excited both the basic scientist and the clinical physician. Therefore, several chapters address these subjects.

It is hoped that this book will not only serve as a resource for understanding the significance of biological studies of the leukemic cell but will also result in a better understanding of the clinical significance of leukemic cell characterization and diagnosis, and therefore be a practical guide for both the laboratorian and clinician. The concepts and studies in acute leukemia are changing so rapidly that our approach to this subject will very likely be different in the next decade. Given the present pace of discoveries, future volumes on acute leukemia will certainly include new concepts regarding past discoveries and will present novel studies to answer current questions. However, we hope that this book will be viewed as a timely resource for understanding the characterization of acute leukemia and the resultant biologic, diagnostic, and therapeutic determinants.

Sanford A. Stass

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1
Introduction
Characterization and Diagnosis of the Acute Leukemias

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I. INTRODUCTION

The acute leukemias are a heterogeneous group of hematopoietic malignancies characterized by a clonal expansion of immature cells. Techniques for cellular characterization have demonstrated heterogeneity beyond the categories of acute lymphoblastic (ALL) and acute nonlymphoblastic leukemia (ANLL), and have led to the recognition of additional subtypes with therapeutic and prognostic significance. The variety of data that can be generated in the process of characterizing leukemic cells is apparent from a reading of the table of contents of this book; what may be less evident is the relative value of the various modalities when applied towards diagnosis, classification, and prognosis. The distinction is not always clear between those tools with practical and established roles in patient management and those of primarily research significance. Certain modalities of clinical value such as determination of the DNA index in childhood ALL (Look et al., 1985) may not be available in every clinical setting. Research techniques used in only a few institutions may prove to have important clinical relevance and eventually become a routine part of the acute leukemia workup. An example of this would be the application of molecular lineage probes in ALL (Korsmeyer et al., 1981, 1983; Arnold et al., 1983; Aisenberg and Wilkes, 1985; Kitchingham et al., 1986). The purpose of this introductory chapter is to place in perspective the characterization and diagnosis of acute leukemia by providing a general overview of some of the topics to be covered later in the book. The chapter will offer general guidelines for application and interpretation of data in acute leukemia.

II. DIAGNOSIS AND PROGNOSIS

A. FAB Classification

The diagnosis and classification of acute leukemia should be based on standard morphologic criteria, best represented at present by the French-American-British (FAB) cooperative group proposals (Bennett et al., 1976, 1981b, 1982, 1985a, 1985b) (see Chapter 2). The advantages of a standardized approach are compelling. The light microscopic examination of the bone marrow including Romanovsky and special cytochemical stains is the standard tool for the diagnosis of acute leukemia. Morphologists require a means to communicate to one another the specific morphologic features identified when rendering a diagnosis; in a larger sense, standardization is crucial to achieve the interinstitutional uniformity necessary to allow meaningful comparison of clinical trials. Most important, a classification system must identify subgroups which have common biologic and clinical features.

In order to be useful, a classification system must be reproducible and have biologic significance and clinical relevance. The reproducibility and prognostic significance of the FAB classification have varied among institutions (Foon et al., 1979; Viana, 1980; Mertelsmann et al., 1980; Miller et al., 1981, 1985; Sultan et al., 1981; Bennett, 1981a; Head et al., 1985; Kalwinsky et al., 1985; van Eys, 1986; Childs, Stass, and Bennett, 1986). Despite the conflicting reports in the literature, we feel the reproducibility is acceptable for routine use if the criteria are closely adhered to and interpreted in a logical fashion. The FAB group has recognized four areas of imprecision in the criteria for the subclassification of acute myelocytic leukemia (AML): the distinction between AML without maturation (FAB-M1) and with maturation (FAB-M2); between FAB-M2 and acute myelomonocytic leukemia (AMML; FAB-M4); between FAB-M4 and acute monocytic leukemia (AMOL; FAB-M5); and between erythroleukemia (FAB-M6) and myelodysplastic syndrome (Bennett, 1985b). The group has recently published revised criteria to address these problems in hopes of improving the performance of the FAB classification (Bennett, 1985b).

The FAB classification identifies subgroups which have common biologic and clinical features. For example, certain subtypes have characteristic cytogenetic abnormalities: t(8;14) in ALL, L3 (Zech, Haglund, and Nilsson, 1976); t(8;21) in FAB-M2 (Rowley, 1973); t(15;17) in acute promyelocytic leukemia (FAB-M3) (Rowley et al., 1977); inversion 16 in acute myelomonocytic leukemia with eosinophilia (FAB-M4 with eosinophilia) (Arthur and Bloomfield, 1983), and abnormalities of 11q in FAB-M5 (Berger et al., 1980). The ALL L3 subtype also has a characteristic B-cell phenotype with detectable surface immunoglobulin. Clinical relevance is demonstrated beyond the important distinction between lymphoid and nonlymphoid lineage, as illustrated by the association of FAB-M3 and FAB-M5 with disseminated intravascular coagulopathy, and extramedullary involvement in FAB-M5.

Despite the conflicting reports concerning prognostic significance, the FAB classification is an important tool in diagnosis and classification. Invariably institutions (and individuals) will tailor the criteria based on personal experience or in response to published exceptions to the rule (Mertelsmann et al., 1980; Head et al., 1985; Miller et al., 1985). This process is unavoidable, but does not necessarily detract from the importance of morphology. However, rational use of the FAB classification requires awareness of the limitations of the criteria in light of present knowledge. The FAB cooperative group has now formally recognized the contribution of cell characterization techniques in the diagnosis of acute megakaryoblastic leukemia (Bennett et al., 1985). We emphasize that the diagnosis and classification of acute leukemia in general is a cumulative effort in which light microscopic findings and cytochemical stains must often be integrated with other parameters such as the electron microscopy, cytogenetics, immunophenotype, flow cytometry, or clinical presentation. These correlates may be essential to arrive at a diagnosis and determine prognosis.

III. ACUTE MYELOCYTIC LEUKEMIA (AML)

A. Diagnosis and Subclassification

The FAB classification is based on the light microscopic appearance of the leukemic cells in the marrow in conjunction with cytochemical stains. We recommend as a minimum a myeloperoxidase (MPO) stain and a nonspecific esterase (NSE) either the α-napthyl butyrate esterase (ANB) or the α-napthyl acetate esterase stain with flouride inhibition (ANA-F). The MPO reaction is positive in myeloblastic leukemia, and is vital for making the distinction from acute lymphoblastic leukemia (ALL). Although Sudan black B has been considered an alternative stain, we feel this is not justified. Sudan black B is not specific for AML, as shown in a review of 350 patients with newly diagnosed ALL, in which six patients (1.6%) were found to have 5% or greater Sudan black positive blasts (Stass et al., 1984). In every newly diagnosed case of acute leukemia, MPO must be performed. As no cytochemical marker exists that is specific for lymphoid lineage commitment, the diagnosis of ALL requires demonstrated absence of myeloperoxidase activity. Some cases of acute leukemia can morphologically resemble lymphoblasts but will show strong MPO positivity and should be classified as AML. In addition, the presence of MPO negative azurophilic cytoplasmic granules has been described in ALL (Grogan et al., 1981; Stein et al., 1983). This emphasizes what we have already noted: cytochemistries should be done in conjunction with morphology prior to rendering a diagnosis. Stains other than MPO, although associated with myeloid lineage, are not as sensitive or specific. Sudan black positivity in ALL has been mentioned. The chloracetate esterase (CAE) generally parallels the MPO stain but may be weakly positive or negative in AML, and punctate positivity can occur in ALL (Keifer, Abromowitch, and Stass, 1985). It is extremely rare for AML to be MPO negative and CAE positive, although we have