9. Cytokines and myeloid-specific genes: Patterns of expression and possible role in proliferation and differentiation of acute myelogenous leukemia cells

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Introduction

Cells from acute myelogenous leukemias (AMLs) are characterized by a block in normal differentiation resulting in dramatically expanded pools of neoplastic precursor cells, both intramedullary and often extramedullary. Other features that also reflect the neoplastic behavior of these cells in vivo include their clonality, the presence of recurrent karyotypic abnormalities, their transplantability, and their often abnormal patterns of expression of and response to cytokines [1]. We are only now beginning to understand some of the mechanisms underlying the observed block in maturation and perpetuation of proliferation of immature myeloid cells in AML. In recent years, molecular biology has provided tools for dissecting the different maturational stages and differentiation pathways of normal myelopoiesis [2]. Also, the study of oncogene activation in primary AMLs and AML cell lines has yielded models of tumorigenesis in this cell system [3]. Similarly, work by a number of laboratories has focused on the possible role of cytokine expression in the pathogenesis of myeloid leukemias. Often, myeloid cell lines established from patients with acute leukemias have served as sources for nucleic acids when isolating myeloid-specific complementary DNAs, and as well-characterized cell populations for in vitro studies. These cultured cell lines in many but not all aspects resemble their primary, uncultured counterparts.

In this chapter, we will try to highlight recent advances made in the understanding of 1) expression of regulation of genes that may be considered markers of the maturational block and 2) the possible role of aberrant cytokine gene expression in stimulation and perpetuation of myeloid leukemic cell growth.

Regulation of myeloid-specific genes in AML

During the normal maturation of white blood cells towards functionally competent, terminally differentiated granulocytic or monocytic cells, the
expression of various genes that code for proteins necessary for phagocyte functions is regulated (reviewed in [2]). This includes surface molecules necessary for cell–cell interactions, receptors for cytokines, and intracellular or secreted proteins crucial for host defense. Many of these genes have been molecularly cloned, and their expression patterns have been studied. It has become apparent that several sets of these genes are co-regulated, and might thus be considered markers of different and distinct maturational steps of normal hematopoiesis. We will focus on features of several members of this somewhat heterogeneous group of genes, their functions, and their patterns of regulation. Thus, we will describe genes whose expression hallmarks either mature myeloblastic/promyelocytic cells (myeloperoxidase, myeloblastin), cells of monoblastic, monocytic/macrophage lineage (lysozyme), and cells that are committed towards maturation to granulocytes (lactoferrin).

Myeloperoxidase

Myeloperoxidase (MPO) is a ~135-kd glycoprotein composed of two 60-kd heavy subunits and two 12-kd light subunits [4]. Mature MPO protein is a cytoplasmic enzyme found in myeloid cells, where it is synthesized and stored in the azurophilic (primary) granules of promyelocytes. MPO catalyzes the reduction of H₂O₂ to yield hypochlorous acid, a potent microbicidal agent, and plays a key role in host defense against various microorganisms [5]. Due to its specificity for myeloid cells, the MPO gene product has been and is one of the most commonly examined marker proteins of white blood cells and thus an important tool when making the diagnosis of myeloid leukemia.

More recently, cDNAs and genomic clones of MPO have been isolated by several laboratories [6–10], and the structure and expression of the MPO gene have been studied. It consists of 12 exons and 11 introns spanning ~13 kb [9,10]. It is localized on the long arm of chromosome 17 (17q22) [11] and probably is not rearranged in acute promyelocytic leukemia (APL), a leukemic syndrome marked by a chromosomal translocation between the long arms of chromosomes 15 and 17, t(15;17) (q22;q11.2) [12,13]. By Northern blot analysis and in situ hybridization, MPO transcripts are readily detectable in cells of the late myeloblastic and promyelocytic stages of differentiation [14,15]. However, these transcripts are absent in normal granulocytes and monocytes in spite of the presence of large amounts of MPO protein, reflecting the high degree of MPO protein stability [14]. In line with this, mRNA rapidly decreases upon induction of granulocytelike or monocytelike differentiation of HL-60 promyelocytic cells after co-cultivation with several different compounds [8,14]. Nuclear run-on analyses and mRNA stability studies show that down-regulation of MPO with terminal myeloid differentiation is controlled predominantly at the transcriptional level [14,16]. However, control may also occur with processing of MPO