

The Process of Leukemogenesis

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Leukemias are monoclonal diseases that arise from cells in the hematopoietic stem and progenitor cell compartment. Consistent with emerging models of carcinogenesis, leukemogenesis is an evolutionary process that involves multiple independent genetic and epigenetic events. Over the last half-century a predominant paradigm has emerged to describe leukemia developing secondary to alkylating drug therapy or exposure to benzene in which progressive dysplastic changes, accompanied by a distinct pattern of clonal cytogenetic abnormalities, give rise to acute myelogenous leukemia. Characterization of these clonal chromosomal aberrations, together with observed alterations in other growth-promoting genes, provides a useful framework for studying chemical leukemogenesis and for use in understanding the origins and development of leukemia in general. — *Environ Health Perspect* 104(Suppl 6):1239–1246 (1996)

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Process of Leukemogenesis

Leukemias are monoclonal diseases that originate from individual cells in the bone marrow. Like other cancers, leukemias often exhibit a progression in their natural history from cells that possess a phenotype exhibiting some remnant of normal differentiation to the progressive loss of maturation ability and the development of an aggressive undifferentiated malignant phenotype (1). A common characteristic of malignant neoplasms, including leukemias, is abnormal regulation of cell growth. A particular challenge to understanding the role of altered regulation of cell growth in the development of leukemia is that differences in the behavior between normal and malignant hematopoietic cells are often subtle. For example, normal hematopoietic progenitor cells possess some characteristics common to malignant cells, among them: the ability to proliferate, survive intravascular transit, and transmigrate

into tissues, and the ability to grow in semisolid media. On the other hand, leukemic cells often do not exhibit total growth-factor independence when first introduced into culture.

Consistent with present models for the origin and progression of neoplasia, leukemia development has long been thought to be a multistep process. Foulds first proposed a multistep progression in which a normal cell must pass through a number of distinct intermediate stages before a frank malignancy develops (2,3). Since that time, a variety of independent observations have emerged to support the conclusion that cancer in general is an evolutionary process in which multiple events involving independent genetic alterations in protooncogenes or suppressor genes together with epigenetic or environmental factors contribute to the development of the full malignant phenotype (4,5). The

best understood example of carcinogenesis in a normal, rapidly proliferating tissue is carcinoma of the colon, in which it appears that as many as five independent genetic and epigenetic changes may be required for the progression of a normal epithelial cell to a carcinoma cell (6,7) (Figure 1). This model is based on the seminal observation by Vogelstein et al. that the incidence of *ras*-gene mutations increased dramatically as a function of the size and malignant phenotype of the tumor and that four molecular alterations accumulated in a manner that paralleled the clinical and histopathological progression of the tumor (7). In addition to illustrating the multifactorial nature of cancer development, these correlative studies reveal that the precise sequence of events is not a constant but that individual tumors can vary in their evolution.

Regulation of Hematopoiesis

A great deal of evidence suggests that protooncogenes and other growth-promoting genes such as those encoding for cytokines or their receptors play an important role in carcinogenesis and malignant transformation. Recent advances in cell and molecular biology have revolutionized our understanding of the regulation of growth in normal hematopoiesis. Therefore, a brief summary of the functions of cytokines in regulating normal hematopoiesis provides a logical foundation for a discussion of the mechanisms of altered cell growth and differentiation occurring in leukemogenesis. Three fundamental cellular processes that define hematopoietic cells are survival, proliferation, and differentiation. The survival and proliferation of hematopoietic progenitor cells (HPC) are controlled by multiple growth factors or cytokines with overlapping functions that act individually or in combination to regulate hematopoiesis (8). Recent studies support the conclusion that either interleukin-3 (IL-3) or granulocyte/macrophage colony-stimulating factor (GM-CSF) are required to sustain the viability of stem cells or early HPC. Early HPC can also be recruited into active cycle in response to these same cytokines or a second stimulus such as *c-kit* ligand (CKL), IL-6, or granulocyte colony-stimulating factor (G-CSF) (9–11). Later committed progenitor cells are controlled by lineage-specific cytokines such as erythropoietin, macrophage colony-stimulating factor (M-CSF), G-CSF, and IL-5. Populations of

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Abbreviations used: AML, acute myelogenous leukemia; β 2AR, β 2-adrenergic receptor; CKL, *c-kit* ligand; CML, chronic myelogenous leukemia; ECGF, endothelial cell growth factor; EGR1, early growth response 1; G-CSF, granulocyte colony-stimulating factor; GM-CSF, granulocyte/macrophage colony-stimulating factor; HPC, hematopoietic progenitor cells; IL, interleukin; M-CSF, macrophage colony-stimulating factor; MDS, myelodysplastic syndrome; PDGFR, platelet-derived growth factor receptor; Rb, retinoblastoma; s, secondary.

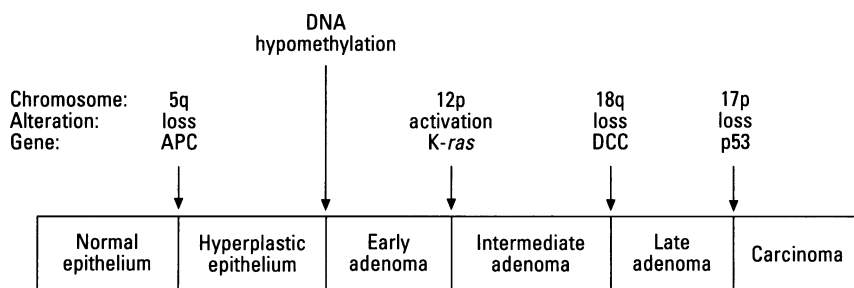


Figure 1. Changes that occur during the evolution of a typical colorectal carcinoma. Schematic representation of a model of tumor progression in which independent steps are required, leading to the activation of at least one protooncogene coupled with the successive loss of several tumor suppressor genes. Adapted from Varmus and Weinberg (6).

early HPC responding to IL-3 and GM-CSF exhibit extensive overlap. Although IL-3 or GM-CSF is required to sustain survival of early dormant progenitor cells (12), IL-3 apparently stimulates cells at an earlier stage of differentiation than GM-CSF (13). Early IL-3 responding populations support the development of T- and B- lymphocyte progeny as well as myeloid and erythroid lineages (14–16). Because IL-3 responsiveness is a characteristic of HPC at multiple levels of differentiation, HPC responsive to GM-CSF may represent a subpopulation of multipotential cells that respond to IL-3 (17). The processes that govern differentiation of HPC are less well understood and are thought by many to be governed via stochastic mechanisms. Nevertheless, it is reasonable to infer that differentiation and lineage commitment are at least indirectly influenced by environmental factors (e.g., cytokines). The role of altered regulation of cytokine expression/response in leukemogenesis is complex, with consistent enhanced expression of GM-CSF or IL-3, resulting in profound myelodysplastic changes (18–20). Altered regulation of clonogenic response to GM-CSF features prominently in both human and murine myeloproliferative disorders and is a frequent early observation in the development of acute myelogenous leukemia (AML) (5,21). Repeated exposure of mice to benzene *in vivo* enhances GM-CSF response (22), and chronic exposure to high concentrations induces a persistent myeloproliferative disorder (23,24). Moreover, the benzene metabolite, hydroquinone, selectively enhances clonogenic response to GM-CSF in murine and human bone marrow cells (25–27). To date, alterations in expression of M-CSF/FMS have not yet been shown to result in abnormal myeloid proliferation.

Relationship between Leukemia, Preleukemia, and Myelodysplastic Syndrome

It is generally recognized that chromosomal aberrations or deletions can alter the regulation and function of protooncogenes and other growth-promoting genes. This, together with our growing knowledge of the function and role of cytokines, their receptors, protooncogenes, and suppressor genes, provides a useful framework for analysis of the respective roles of altered cell growth and differentiation in chemical leukemogenesis. To this end, an impressive literature exists that describes the natural history of leukemia secondary to alkylating drug or occupational exposure in which the development of AML is *a*) preceded by progressive dysplastic or “preleukemic” changes, and *b*) accompanied by a distinct pattern of clonal cytogenetic abnormalities.

The concept of preleukemia originated around the turn of the century with the observation that AML could be preceded by a cytopenic dysplasia involving one or more hematopoietic cell lines (28). However, preleukemia was not generally accepted as a clinical entity until the widespread introduction of radiation and chemotherapy in the treatment of other types of cancer. It is now widely recognized that persistent cytopenias and other blood dyscrasias, including dyserythropoiesis, dysgranulopoiesis, and dysmegakaryopoiesis, frequently precede the onset of leukemia in patients developing AML secondary to exposure to benzene or alkylating agents (1,29–32). Over the past decade the term preleukemia has been largely supplanted by the more functional classification of the myelodysplastic syndromes (MDS) by the fast atom bombardment (FAB) (33). Differences in growth and differentiation between MDS and

AML find analogy in the progression of solid tissue tumors from metaplasia and dysplasia to carcinoma. This observation, together with the frequent progression of secondary s-MDS to frank AML, leads to the inescapable conclusion that MDS and AML should be considered a single disease continuum when viewed in the context of chemical leukemogenesis.

As a secondary malignancy, AML clearly dominates the leukemia literature. At present, the world literature contains approximately 38,000 cases of individuals treated with alkylating agents or radiation for primary malignancies or immune disorders that have been followed for development of secondary malignancies, the most prominent of which is AML. These typically involve M1, M2, M4, M6, but not M3 or M5 subtypes of AML, based on the FAB classification system (34,35). From these studies at least 1322 cases of AML and 320 cases of MDS have been reported (29,30,36–57). If one includes all cases attributed to solvent exposure in general or benzene specifically, whether anecdotal or reported in case-control or cohort studies, the total number of secondary leukemias approaches 2100, over 96% of which are MDS/AML (44,58–70). The frequency of secondary AML/MDS varies markedly depending on individual therapeutic regimen but historically has ranged between 0.6 and 17%, with relative risks averaging about 100-fold (range 9–320 X). These studies establish a consistent and very strong pattern in which the development of AML is preceded by a period of preleukemia in 33 to 80% of the cases and is accompanied by clonal cytogenetic abnormalities involving loss of all or part of chromosomes 5 and 7 (Table 1). On average the frequency of deletions or loss of chromosomes 5 and 7 in studies of patients who develop MDS or AML after antineoplastic therapy ranges between 85 and 95% (71–76). The same cytogenetic abnormalities occur much less frequently in *de novo* AML: in 660 *de novo* cases chromosomes 5 and 7 were observed in 4.2 and 4.4%, respectively, and simultaneously in 3.2% (77,78). Consistent with the evolution of s-AML described previously, most patients presenting with clonal chromosomal aberrations involving the loss of all or part of chromosomes 5 or 7 exhibit a preleukemic phase prior to the onset of AML (79). Analysis of the specific pattern of cytogenetic involvement in AML developing secondary to benzene exposure is complicated by the ambiguities commonly associated with the characterization of

Table 1. Cytogenetic characteristics of secondary acute myelogenous leukemia.

Study	Clonal aberrations		-5, -7, 5q-, 7q- or both	
	<i>De novo</i> AML, %	Secondary AML, %	<i>De novo</i> AML, %	Secondary AML, %
FIWCL (77)	54	73	12	48
Le Beau et al. (73)	56	97	16	87
Rowley et al. (72)	—	96	—	92
Rowley et al. (71)	—	100	—	90
Pedersen-Bjergaard et al. (121)	—	86	—	89
Wald and Conner (122)	—	87	—	80
Mitelman et al. (59)	24	83	12	84
Golomb et al. (60)	—	75	—	67
Fagioli et al. (61)	29	88	20	46

exposure in occupational and retrospective studies. Issues related to the specificity and intensity of benzene exposure notwithstanding, the classic pattern involving a high frequency of loss of all or part of chromosomes 5 and/or 7 is also observed in studies of patients occupationally exposed to benzene specifically or solvents among which benzene is the only recognized leukemogen (58–61,63,65,80,81). The consistency of this pattern is all the more impressive when one considers that the association between exposure and cytogenetic abnormalities in occupational studies is invariably diluted; i.e., studies providing the greatest detail on cytogenetic abnormalities are weakest in the characterization of exposure criteria. Independently, recent studies of peripheral lymphocytes in benzene-exposed individuals in China have shown that benzene exposure induces aneuploidy of C-group chromosomes, with an especially strong effect on chromosome 7. Both hyper- and hypodiploidy of chromosome 7 occurred more frequently in benzene-exposed workers than in matched controls (82). Other nonrandom clonal chromosomal abnormalities, such as +8 or +21, have been observed to be increased in either exposed or nonexposed populations, depending on the study (58–60,62,81). These observations suggest that although some chromosomes other than 5 or 7 can be involved in the pathogenesis of AML secondary to exposure to benzene or chemotherapeutic alkylating agents, they are not useful in discriminating between *de novo* and s-AML (83). More recently, a distinct pattern of secondary AML has been observed following therapy with drugs targeting DNA-topoisomerase II. The pattern of AML developing as a consequence of exposure to these agents, examples of which include etoposide and teniposide, includes the notable absence of a preleukemic phase, frequent presentation of an M3 subtype, and balanced chromosome

aberrations involving bands 11q23 and 21q22 (84). Although it has been hypothesized that benzene metabolites may interfere with topoisomerase II (85), the pattern of leukemias and chromosomal aberrations typically associated with inhibition of this enzyme has yet to be observed in occupationally exposed populations.

5-/5q-: A Model for the Pathogenesis of Leukemia

The pattern of reoccurring chromosomal abnormalities associated with the development of leukemia can be used as a guide in understanding the etiology and pathogenesis

of these diseases. A number of gene loci have been mapped to chromosome 7; however, their function(s) remains largely unknown (86). However, recent progress in the mapping of genes to the region of chromosome 5 involved in s-MDS/AML, provides a logical starting point for a discussion of possible mechanisms of leukemogenesis. Deletions of all or part of chromosomes 5 or 7 are the earliest clonal alterations that have been detected in MDS/AML. Deletions associated with 5q- in s-MDS/AML are usually interstitial without translocation of the deleted material (73,87). The variability of the breakpoints, together with identification of the critical region (5q31), suggests that the relevant genetic event may be the deletion of a critical gene sequence rather than consistent juxtaposition of DNA sequences that may occur in chromosome translocation (Figure 2) (73). A cluster of genes involved in the regulation of hematopoiesis is located at q31 on chromosome 5 (Figure 3). These include GM-CSF, IL-3, IL-4, IL-5, CD14 (which encodes a myeloid-specific surface molecule that has structural characteristics of a receptor), and early growth response 1 (EGR1) (an *EGR* gene with *fos*-like properties) (88).

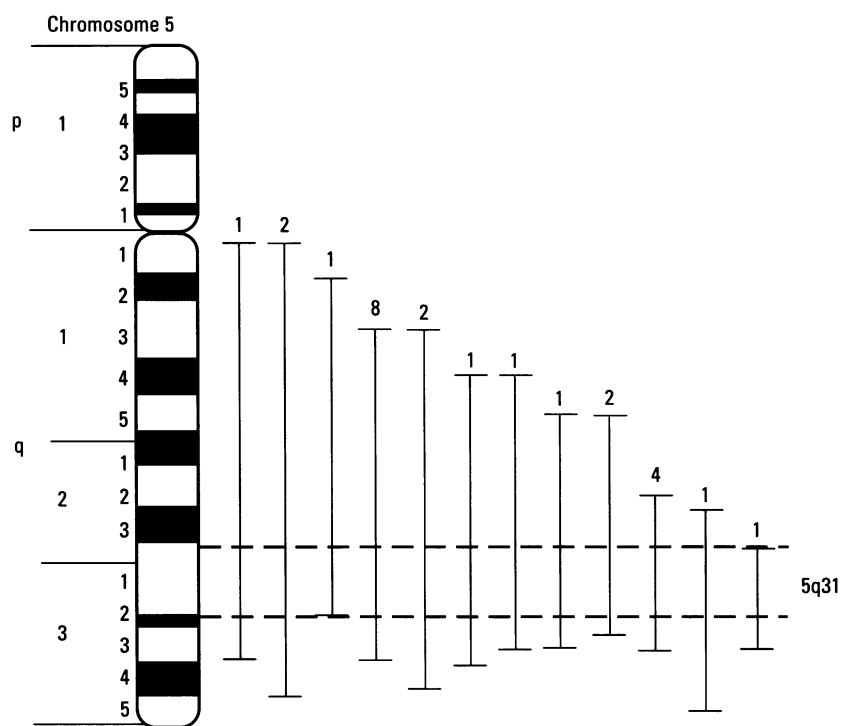


Figure 2. Fragile site on chromosome 5 associated with region of critical deletion in patients with t-AML. Vertical bars indicate deleted segments and numbers indicate number of patients with same deletion. Dashed bars indicate smallest overlapping region of 5q31. Reproduced from Rowley and Le Beau (88), with permission.

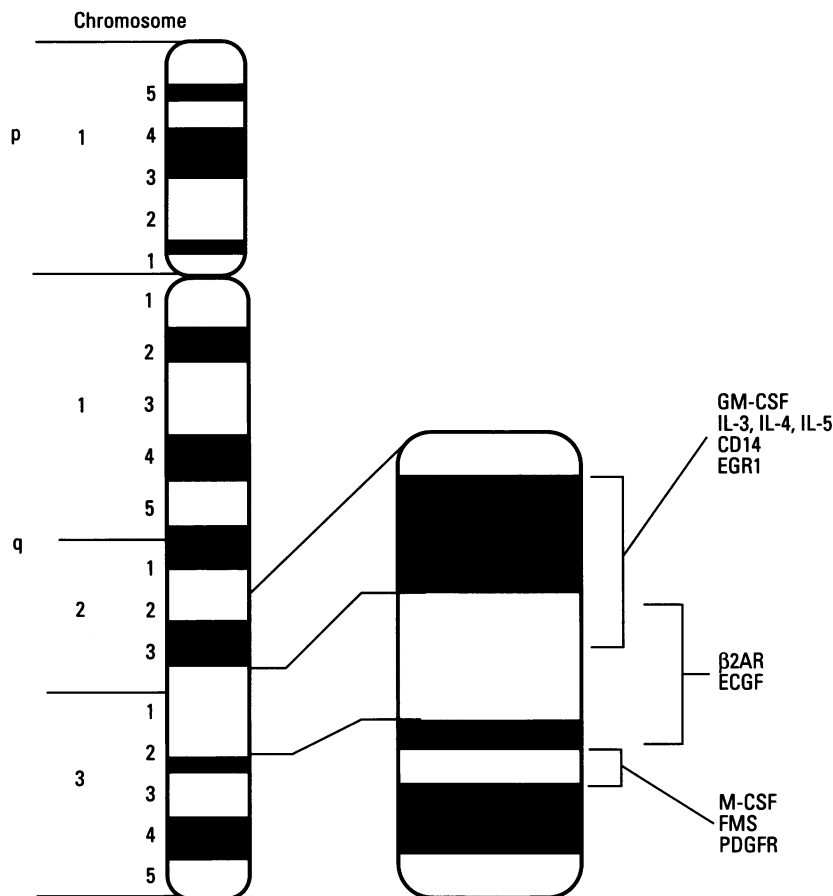


Figure 3. Genetic map of genes located in and adjacent to the 5q31-critical region deleted in patients with *t*-AML. Modified from Rowley and Le Beau (88). Abbreviations: β 2AR, β 2-adrenergic receptor; ECGF, endothelial cell growth factor; PDGFR, platelet-derived growth factor receptor.

There is no evidence for homozygous deletion of any of these genes in AML, such as has been described for retinoblastoma in which the absence of one allele of retinoblastoma gene is followed by a second somatic mutation, resulting in loss of both copies of the gene (89). The most consistent deletion associated with 5q31 appears to involve the loss of one allele encoding either GM-CSF or EGR1 (88). Close to, but lying outside of, the critical region is the protooncogene, *c-fms*, the receptor for M-CSF that acts exclusively on cells of monomacrophage lineage (90). Ostensibly, a critical role for *c-fms* in the development of AML is an attractive hypothesis in that it encodes a cytokine receptor. However, the weight of evidence suggests that *c-fms* is probably not involved in early events in AML development. M-CSF and *c-fms* lie outside the critical deletion region at 5q31 (88), and although FMS expression can be detected on leukemia cells in approximately 30% of

AMLs, abnormal myeloid proliferation has yet to be demonstrated as a result of inappropriate expression of M-CSF or FMS (5). Nevertheless, the incidence of point mutations in the *c-fms* gene in AML or MDS has been reported to be 13% in one series, suggesting that subtle changes in *c-fms* expression may contribute in some way to the development of the malignant phenotype (91).

At present, there are at least two molecular hypotheses for the role of 5q- abnormalities in the development of MDS/AML. First is the loss of a single allele of a heretofore unidentified tumor suppressor gene that results in production of a nonfunctional protein; this would be analogous to the inactivation of p53 by point mutation. The second hypothesis is that loss of a single allele leads to altered gene dosage and a reduction in the level of gene product, such as GM-CSF. A third possibility is that involvement of intact genes adjacent to the interstitial deletion cannot at this

time be entirely excluded since their function could ostensibly be influenced by the structural changes that have occurred in the chromosome.

In addition to early involvement of genes on chromosome 5, a growing number of protooncogenes have been reported to undergo structural or functional alterations in AML with widely differing frequency. These have been the subject of a number of reviews (5,92-94). To date, no single or consistent pattern of protooncogene involvement has been associated with AML development, suggesting that multiple genes may interact via different pathways in the evolution of the disease. This discussion will focus only on a small number of these genes. The human *ras* genes encode p21 proteins that appear to play an important role as second messengers in tyrosine kinase receptor-mediated signal transduction (95). It has been hypothesized that *ras* mutations may feature prominently in the development of s-AML (96). However, what role *ras* mutations play in the development of either *de novo* or s-AML remains uncertain. In a large number of studies, *ras* protooncogene activation, principally N-RAS, has been variously reported to occur in about 25% of cases of *de novo* AML (92,95). However, clonal chromosomal aberrations have been demonstrated to precede *ras* involvement in the evolution of the disease (97,98) and, when identified, *ras* mutations tend to be present only in subclones of the leukemic cells (97,99). To date, RAS involvement has not proven to be either a prognostic indicator or to correlate with FAB subtype. From these studies it appears that *ras* mutation is an unlikely early event in AML and is insufficient to cause the disease. However, *ras* activation can occur at a number of stages in the development of AML and it cannot be excluded that RAS may play a role in the evolution or progression of at least some cases of AML (100). The p53 gene encodes a phosphoprotein nuclear transcription factor that is spatially regulated within the cell during the cell cycle. The wild-type protein is known to limit cell growth apparently by two independent mechanisms: mediating apoptosis and as a checkpoint, regulating the length of *G1* (101). Often referred to as a tumor suppressor gene, inactivation of p53 is an important event in the transformation of many tumors and is the most frequently encountered gene mutation in human cancer (102). Inactivation of p53 is involved in the progression to blast crisis in

20 to 30% of chronic myelogenous leukemia (CML) cases, but it is only encountered in a few percent of MDS/AML cases (103). Within this small subset of AMLs, p53 inactivation is a late event and is apparently associated with loss of a differentiated cell phenotype, aggressive course, and a poor prognosis. The retinoblastoma (Rb) gene was the first identified tumor suppressor gene and like p53 encodes a phosphonuclear protein that is involved in regulating critical events in the cell cycle (104,105). Absent or decreased expression of Rb protein is observed in approximately 30% of AML cases and is associated with a particularly dismal response to therapy (106,107).

A Model for Leukemogenesis

Up to this point the discussion has focused exclusively on the role of clonal hematopoietic stem and progenitor cell abnormalities in the development and evolution of MDS/AML. Another hypothesis has been proposed in which altered growth factor production by fibroblasts, endothelial cells, and macrophages may feature prominently in the development of leukemias (108). As the basis for a stand-alone theory of leukemogenesis, each of these alternatives possesses individual strengths and weaknesses. A diverse set of observations argues persuasively that the ultimate clonal derivation of most cases of AML and MDS are HPC essentially restricted to the myeloid lineage (109–112). Superficially, a clonal lesion would appear to be incompatible with the notion that persistent and progressive myeloproliferative disease could arise as a consequence of altered microenvironmental influences that are ostensibly independent and oligoclonal. On the other hand, how allelic deletion of a cytokine gene such as GM-CSF in a single clone of progenitor cells can predispose to a series of events that ultimately leads to AML remains a nagging enigma. The basis for the argument for a stromal origin is that multiple peripheral cytopenias and dysplasias are the rule rather

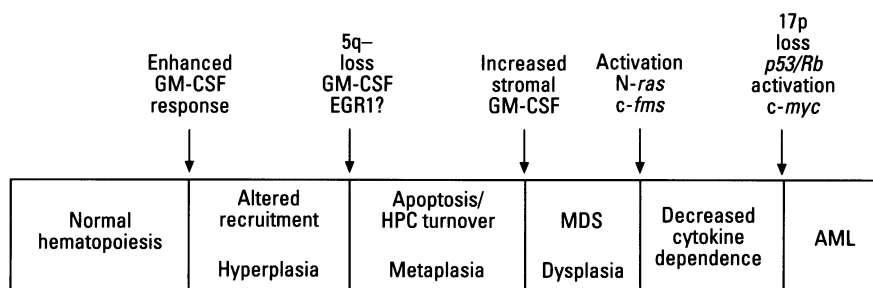


Figure 4. Hypothetical model for the evolution of secondary leukemia involving 5q-. Schematic representation of a model of abnormal myeloproliferative progression in which early events, including alterations in cytokine response and loss of heterozygosity (5q-), are followed by activation of at least one protooncogene coupled with the successive loss of a tumor suppressor gene.

than the exception in the evolution of s-AML, and abnormal cytokine production is frequently encountered in cases of MDS/AML (113–116).

Alternatively, one can propose an integrative model of leukemogenesis that is compatible with both clonal and microenvironmental involvement in the development and progression of these myeloproliferative disorders. In hematopoiesis, there is considerable evidence that cell viability and cell growth are functions that can be dissociated (117). Raza et al. (118,119) have recently reported an increase in both cell turnover and apoptosis in bone marrow of patients with MDS relative to normal bone marrow that is in sharp contrast to AML, in which proliferation is high but the frequency of apoptotic cells is low. A chromosomal aberration in an early progenitor cell could result in allelic deletion of one GM-CSF gene, resulting in a commensurate decrease in intracellular GM-CSF. Using antisense technology, Pech and co-workers provide evidence that low-level autocrine regulation of GM-CSF may regulate survival in early progenitor cells independent of exogenous GM-CSF that is associated with proliferation (120). If intracellular GM-CSF were insufficient to suppress apoptosis in maturing cells of the affected clone, a corresponding increase in

exogenous GM-CSF occurring in physiologic response to increased apoptosis would drive cell proliferation, leading to an increase in overall cell turnover in the affected clone. This model is consistent with the regulatory paradox in MDS in which bone marrow hyperplasia is accompanied by ineffective hematopoiesis and cytopenias. Subsequent events involving genes linked directly or indirectly to cell survival and maturation (e.g., *ras*, *p53*, or *Rb*) could enable the escape of the subclone from the apoptotic treadmill and development of a frank AML (Figure 4).

This model is only one possible explanation for the origins and progression of leukemia that is compatible with roles for both clonal and microenvironmental events. The earliest observations associated with the development of MDS/AML suggest that a relatively small number of alternative events predispose the development of leukemia. These most likely involve a clonal chromosomal abnormality together with altered regulation of cytokine response. However, the diversity of gene involvement in later stages of AML progression is consistent with an emerging pattern in cancer biology in which multiple alternative genetic pathways converge in the development of a specific tumor type.

REFERENCES

1. Foucar K, McKenna RW, Bloomfield CD, Bowers TK, Brunning RD. Therapy-related leukemia. *Cancer* 43:1285–1296 (1979).
2. Foulds L. Neoplastic Development. New York:Academic Press, 1969.
3. Foulds L. The experimental study of tumor progression: a review. *Cancer Res* 14:327–339 (1954).
4. Land H, Parada LF, Weinberg RA. Tumorigenic conversion of primary embryo fibroblasts requires at least two cooperating oncogenes. *Nature* 304:596–602 (1983).
5. Willman CL, Whittaker MH. The molecular biology of acute myeloid leukemia. Proto-oncogene expression and function in normal and neoplastic myeloid cells. *Clin Lab Med* 10:769–796 (1990).
6. Varmus H, Weinberg RA. Genes and the Biology of Cancer. New York:Scientific American Library, 1993.
7. Vogelstein B, Fearon ER, Hamilton SR, Kern SE, Preisinger AC, Leppert M, Nakamura Y, White R, Smits AMM, Bos JL. Genetic alterations during colorectal-tumor development. *N Engl J Med* 319(9):525–532 (1988).

8. Leary AG, Zeng HQ, Clark SC, Ogawa M. Growth factor requirements for survival in G₀ and entry into the cell cycle of primitive human hemopoietic progenitors. *Proc Natl Acad Sci USA* 89:4013-4017 (1992).
9. Ikebuchi K, Clark SC, Ihle JN, Souza LM, Ogawa M. Granulocyte colony-stimulating factor enhances interleukin 3-dependent proliferation of multipotential hemopoietic progenitors. *Proc Natl Acad Sci USA* 85:3445-3449 (1988).
10. Ogawa M, Matsuzaki Y, Nishikawa S, Hayashi S-I, Kunisada T, Sudo T, Kina T, Nakauchi H, Nishikawa S-I. Expression and function of *c-kit* in hemopoietic progenitor cells. *J Exp Med* 174:63-71 (1991).
11. Bodine DM, Orlic D, Birkett NC, Seidel NE, Zsebo KM. Stem cell factor increases colony-forming unit-spleen number *in vitro* in synergy with interleukin-6, and *in vivo* in *SL/Sl^d* mice as a single factor. *Blood* 79:913-919 (1992).
12. Gibson FM, Bagnara M, Ioannidou E, Gordon-Smith EC. Interaction of granulocyte-macrophage colony-stimulating factor and interleukin-3 in human long-term bone marrow culture. *Exp Hematol* 20:235-240 (1992).
13. Leary GA, Yang YC, Clark SC, Gasson JC, Golde DW, Ogawa M. Recombinant gibbon interleukin-3 supports formation of human multilineage colonies and blast cell colonies in culture: comparison with recombinant human granulocyte-macrophage colony-stimulating factor. *Blood* 70:1343-1348 (1987).
14. Tsunoda J-I, Okada S, Suda J, Nagayoshi K, Nakauchi H, Hatake K, Miura Y, Suda T. *In vivo* stem cell function of interleukin-3-induced blast cells. *Blood* 78:318-322 (1991).
15. Wood PMD, Jordan RK, Givan AL, Brooks CG. IL-3 and IL-4 affect thymocyte differentiation in organ culture. *Immunology* 71:83-89 (1990).
16. Soloff RS, Dempsey D, Jennings SR, Wolcott RM, Chervenak R. Characterization of the progeny of pre-T cells maintained *in vitro* by IL-3: appearance in the periphery and Vb utilization *in vivo*. *Cell Immunol* 135:132-142 (1991).
17. Koike K, Ogawa M, Ihle JN, Miyake T, Shimizu T, Miyajima A, Yokota T, Arai K-I. Recombinant murine granulocyte-macrophage (GM) colony-stimulating factor supports formation of GM and multipotential blast cell colonies in culture: comparison with the effects of interleukin-3. *J Cell Physiol* 131:458-464 (1987).
18. Chang JM, Metcalf D, Lang RA, Gonda TJ, Johnson GR. Nonneoplastic hematopoietic myeloproliferative syndrome induced by dysregulated multi-CSF (IL-3) expression. *Blood* 73:1487-1497 (1989).
19. Lang RA, Metcalf D, Gough NM, Dunn AR, Gonda TJ. Expression of a hemopoietic growth factor cDNA in a factor-dependent cell line results in autonomous growth and tumorigenicity. *Cell* 43:531-542 (1985).
20. Stocking C, Loliger C, Kawai M, Suciu S, Gough N, Ostertag W. Identification of genes involved in growth autonomy of hematopoietic cells by analysis of factor-independent mutants. *Cell* 53:869-879 (1988).
21. Young KM. Myeloproliferative disorders. *Vet Clin N Am: Small Anim Prac* 15:769-781 (1985).
22. Dempster AM, Snyder CA. Short term benzene exposure provides a growth advantage for granulopoietic progenitor cells over erythroid progenitor cells. *Arch Toxicol* 64:539-544 (1990).
23. Farris GM, Everitt JI, Irons RD, Popp JA. Carcinogenicity of inhaled benzene in CBA mice. *Fundam Appl Toxicol* 20:503-507 (1993).
24. Cronkite EP, Drew RT, Inoue T, Hirabayashi Y, Bullis JE. Hematotoxicity and carcinogenicity of inhaled benzene. *Environ Health Perspect* 82:97-108 (1989).
25. Irons RD, Stillman WS, Colagiovanni DB, Henry VA. Synergistic action of the benzene metabolite hydroquinone on myelopoeitic stimulating activity of granulocyte/macrophage colony-stimulating factor *in vitro*. *Proc Natl Acad Sci USA* 89:3691-3695 (1992).
26. Irons RD, Stillman WS. Cell proliferation and differentiation in chemical leukemogenesis. *Stem Cells* 11:235-242 (1993).
27. Irons RD, Stillman WS. Impact of benzene metabolites on differentiation of bone marrow progenitor cells. In: *Benzene '95: International Conference on the Toxicity, Carcinogenesis, and Epidemiology of Benzene*, 17-20 July 1995, Piscataway, NJ. Abstract.
28. Layton DM, Mufti GJ. Myelodysplastic syndromes: their history, evolution and relation to acute myeloid leukaemia. *Blut* 53:423-436 (1986).
29. Casciato DA, Scott JL. Acute leukemia following prolonged cytotoxic agent therapy. *Medicine* 58(1):32-47 (1979).
30. Anderson RL, Bagby GC Jr, Richert-Boe K, Magenis RE, Koler RD. Therapy-related preleukemic syndrome. *Cancer* 47:1867-1871 (1981).
31. Coltman CA, Dahlberg S. Treatment-related leukemia. *N Engl J Med* 322:52-53 (1990).
32. Advani SH, Doval DC, Gopal R, Nair CN, Kutty PM. Therapy-related leukemia. *Oncology* 40:268-272 (1983).
33. Bennett JM, Catovsky D, Daniel MT, Flandrin G, Galton DAG, Gralnick HR, Sultan C. Proposals for the classification of the myelodysplastic syndromes. *Br J Haematol* 51:189-199 (1982).
34. Bennett JM, Catovsky D, Daniel MT, Flandrin G, Galton DAG, Gralnick HR, Sultan C. Proposed revised criteria for the classification of acute myeloid leukemia. *Ann Intern Med* 103:626-629 (1985).
35. Levine EG, Bloomfield CD. Leukemias and myelodysplastic syndromes secondary to drug, radiation, and environmental exposure. *Semin Oncol* 19(1):47-84 (1992).
36. Allan WSA. Acute myeloid leukaemia after treatment with cytostatic agents. *Lancet* October 10:775 (1970).
37. Bartolucci AA, Liu C, Durant JR, Gams RA. Acute myelogenous leukemia as a second malignant neoplasm following the successful treatment of advanced Hodgkin's disease. *Cancer* 52:2209-2213 (1983).
38. Boivin J-F, Hutchison GB. Leukemia and other cancers after radiotherapy and chemotherapy for Hodgkin's disease. *J Natl Cancer Inst* 67(4):751-760 (1981).
39. Coleman CN, Williams CJ, Flint A, Glatstein EJ, Rosenberg SA, Kaplan HS. Hematologic neoplasia in patients treated for Hodgkin's disease. *N Engl J Med* 297:1249-1252 (1977).
40. Devereux S. Therapy associated leukaemia. *Blood Rev* 5:138-145 (1991).
41. Greene MH, Harris EL, Gershenson DM, Malkasian GD Jr, Melton LJ III, Dembo AJ, Bennett JM, Moloney WC, Boice JD Jr. Melphalan may be a more potent leukemogen than cyclophosphamide. *Ann Intern Med* 105:360-367 (1986).
42. Grunwald HW, Rosner F. Acute myeloid leukemia following treatment of Hodgkin's disease: a review. *Cancer* 50:676-683 (1982).
43. Grunwald HW, Rosner F. Acute leukemia and immunosuppressive drug use: a review of patients undergoing immunosuppressive therapy for non-neoplastic diseases. *Arch Intern Med* 139:461-466 (1979).
44. Hoppe RT. Secondary leukemia and myelodysplastic syndrome after treatment for Hodgkin's disease. *Leukemia* 6 (Suppl 4):155-157 (1992).
45. Kantarjian HM, Keating MJ, Walters RS, Smith TL, Cork A, McCredie KB, Freireich EJ. Therapy-related leukemia and myelodysplastic syndrome: clinical, cytogenetic, and prognostic features. *J Clin Oncol* 14(12):1748-1757 (1986).
46. Kyle RA, Gertz MA. Second malignancies after chemotherapy. In: *The Chemotherapy Source Book* (Perry MC, ed). Baltimore: Williams & Wilkins, 1992; 689-702.
47. Kyle RA. Second malignancies associated with chemotherapeutic agents. *Semin Oncol* 9(1):131-142 (1982).
48. Lishner M, Slingerland J, Barr J, Panzarella T, Degendorfer P, Sutcliffe S. Second malignant neoplasms in patients with non-Hodgkin's lymphoma. *Hematol Oncol* 9:169-179 (1991).
49. Pedersen-Bjergaard J, Larsen SO. Incidence of acute nonlymphocytic leukemia, preleukemia, and acute myeloproliferative syndrome up to 10 years after treatment of Hodgkin's disease. *N Engl J Med* 307:965-971 (1982).

50. Pedersen-Bjergaard J. Radiotherapy- and chemotherapy-induced myelodysplasia and acute myeloid leukemia. A review. *Leuk Res* 16:61–65 (1992).
51. Pedersen-Bjergaard J, Nissen NI, Sorensen HM, Hou-Jensen K, Larsen MS, Ernst P, Ersbol J, Knudtzon S, Rose C. Acute non-lymphocytic leukemia in patients with ovarian carcinoma following long-term treatment with treosulfan (=dihydroxybusulfan). *Cancer* 45:19–29 (1980).
52. Rosner F, Grunwald HW. Hodgkin's disease and acute leukemia: report of eight cases and review of the literature. *Am J Med* 58:339–353 (1975).
53. Rosner F, Grunwald HW. Multiple myeloma terminating in acute leukemia. *Am J Med* 57:927–939 (1974).
54. Kaldor JM, Day NE, Pettersson F, Clarke EA, Pedersen D, Mehnert W, Bell J, Host H, Prior P, Karjalainen S, Neal F, Koch M, Band P, Choi W, Kirn VP, Arslan A, Zaren B, Belch AR, Storm H, Kittelmann B, Fraser P, Stovall M. Leukemia following chemotherapy for ovarian cancer. *N Engl J Med* 322:1–6 (1990).
55. Smit CGS, Meyler L. Acute myeloid leukaemia after treatment with cytostatic agents. *Lancet* September 26:671–672 (1970).
56. Trump DL, Cowall DE. Acute myelogenous leukemia as a late complication of the multimodality therapy for Hodgkin's disease. *Johns Hopkins Med J* 141:249–251 (1977).
57. Tucker MA, Coleman CN, Cox RS, Varghese A, Rosenberg SA. Risk of second cancers after treatment for Hodgkin's disease. *N Engl J Med* 318(2):76–81 (1988).
58. Mitelman F, Nilsson PG, Brandt L, Alimena G, Gastaldi R, Dallapiccola B. Chromosome pattern, occupation, and clinical features in patients with acute nonlymphocytic leukemia. *Cancer Genet Cytogenet* 4:197–214 (1981).
59. Mitelman F, Brandt L, Nilsson PG. Relation among occupational exposure to potential mutagenic/carcinogenic agents, clinical findings, and bone marrow chromosomes in acute nonlymphocytic leukemia. *Blood* 52:1229–1237 (1978).
60. Golomb HM, Alimena G, Rowley JD, Vardiman JW, Testa JR, Sovik C. Correlation of occupation and karyotype in adults with acute nonlymphocytic leukemia. *Blood* 60:404–411 (1982).
61. Fagioli F, Cuneo A, Piva N, Carli MG, Previati R, Balboni M, Tomasi P, Cariani D, Scapoli G, Castoldi G. Distinct cytogenetic and clinicopathologic features in acute myeloid leukemia after occupational exposure to pesticides and organic solvents. *Cancer* 70:77–85 (1992).
62. Crane MM, Keaing MJ, Trujillo JM, Labarthe DR, Frankowski RF. Environmental exposures in cytogenetically defined subsets of acute nonlymphocytic leukemia. *JAMA* 262:634–6639 (1989).
63. Narod SA, Dube ID. Occupational history and involvement of chromosomes 5 and 7 in acute nonlymphocytic leukemia. *Cancer Genet Cytogenet* 38:261–269 (1989).
64. Michels SD, McKenna RW, Arthur DC, Brunning RD. Therapy-related acute myeloid leukemia and myelodysplastic syndrome: a clinical and morphologic study of 65 cases. *Blood* 65(6):1364–1372 (1985).
65. Cuneo A, Fagioli F, Pazzi I, Tallarico A, Previati R, Piva N, Carli GM, Balboni M, Castoldi G. Morphologic, immunologic and cytogenetic studies in acute myeloid leukemia following occupational exposure to pesticides and organic solvents. *Leuk Res* 16:789–796 (1992).
66. Miller JS, Arthur DC, Litz CE, Neglia JP, Miller WJ, Weisdorf DJ. Myelodysplastic syndrome after autologous bone marrow transplantation: an additional late complication of curative cancer therapy. *Blood* 83:3780–3786 (1994).
67. Aksoy M. *Benzene Carcinogenicity*. Boca Raton, FL: CRC Press, 1988.
68. Vigliani EC, Forni A. Benzene and leukemia. *Environ Res* 11:122–127 (1976).
69. Forni A, Vigliani EC. Chemical leukemogenesis in man. *Semin Hematol* 7(2):211–223 (1974).
70. Decoufle P, Blattner WA, Blair A. Mortality among chemical workers exposed to benzene and other agents. *Environ Res* 30:16–25 (1983). Abstract.
71. Rowley JD, Golomb HM, Vardiman J. Nonrandom chromosomal abnormalities in acute nonlymphocytic leukemia in patients treated for Hodgkins disease and non-Hodgkins lymphomas. *Blood* 50:759–770 (1977).
72. Rowley JD, Golomb HM, Vandiman JW. Nonrandom chromosome abnormalities in acute leukemia and dysmyelopoietic syndromes in patients with previously treated malignant disease. *Blood* 58:759–767 (1981).
73. Le Beau MM, Albain KS, Larson RA, Vardiman JW, Davis EM, Blough RR, Golomb HM, Rowley JD. Clinical and cytogenetic correlations in 63 patients with therapy-related myelodysplastic syndromes and acute nonlymphocytic leukemia: further evidence for characteristic abnormalities of chromosomes no. 5 and 7. *J Clin Oncol* 4(3):325–345 (1986).
74. Pedersen-Bjergaard J, Philip P, Pedersen NT, Hou-Jensen K, Sveigaard A, Jensen G, Nissen NI. Acute nonlymphocytic leukemia, preleukemia, and acute myeloproliferative syndrome secondary to treatment of other malignant diseases. *Cancer* 54:452–462 (1984).
75. Johansson B, Mertens F, Heim S, Kristoffersson U, Mitelman F. Cytogenetics of secondary myelodysplasia (sMDS) and acute nonlymphocytic leukemia (sANLL). *Eur J Haematol* 47:17–27 (1991).
76. Bitter MA, Le Beau MM, Rowley JD, Larson RA, Golomb HM, Vardiman JW. Associations between morphology, karyotype, and clinical features in myeloid leukemias. *Hum Pathol* 18:211–225 (1987).
77. Fourth International Workshop on Chromosomes in Leukemia 1982. The correlation of karyotype and occupational exposure to potential mutagenic/carcinogenic agents in acute nonlymphocytic leukemia. *Cancer Genet Cytogenet* 11:326–331 (1984).
78. Bloomfield CD, Goldman A, Hossfeld D, de la Chapelle A. Clinical significance of chromosomal abnormalities in acute nonlymphoblastic leukemia. *Cancer Genet Cytogenet* 11:332–350 (1984).
79. Pedersen-Bjergaard J, Philip P, Mortensen BT, Ersboll J, Jensen G, Panduro J, Thomsen M. Acute nonlymphocytic leukemia, preleukemia, and acute myeloproliferative syndrome secondary to treatment of other malignant diseases. Clinical and cytogenetic characteristics and results of *in vitro* culture of bone marrow and HLA typing. *Blood* 57(4):712–723 (1981).
80. Smith MT, Zhang L, Rothman N, Wang Y, Hayes RB, Li GL, Yin SN. Interphase cytogenetics of workers exposed to benzene. *Toxicologist* 15:87 (1995). Abstract.
81. Van den Berghe H, Louwagie A, Broeckaert-Van Orshoven A, David G, Verwilghen R. Chromosome analysis in two unusual malignant blood disorders presumably induced by benzene. *Blood* 53:558–566 (1979).
82. Zhang L, Rothman N, Wang Y, Hayes RB, Yin S-N, Holland N, Dosemeci M, Wang Y-Z, Kolachana P, Xi L-Q, Li G-L, Smith MT. Aneuploidy of chromosomes 7, 8 and 9 detected by fluorescence *in situ* hybridization in workers exposed to benzene. *Proc Natl Acad Sci USA* 92:4069–4073 (1995). (Abstract).
83. Le Beau MM, Rowley JD. Chromosomal abnormalities in leukemia and lymphoma: clinical and biological significance. *Adv Hum Genet* 15:1–54 (1986).
84. Stone R, Neuberg D, Soiffer R, Whelan M, Leavitt P, Aster J, Takvorian R, Rabinow S, Ritz J, Freedman A, Nadler L. Myelodysplastic syndrome (MDS) as a complication after autologous bone marrow transplantation (ABMT) for non Hodgkin's lymphoma (NHL). *Blood* 82:196a (1993). Abstract.
85. Chen HW, Frantz C, Eastmond DA. Inhibition of topoisomerase enzymes by phenolic metabolites: a potential mechanism for benzene's clastogenic effects. In: *Benzene '95: International Conference on the Toxicity, Carcinogenesis, and Epidemiology of Benzene*, 17–20 July 1995, Piscataway, NJ. Abstract.
86. Luna-Fineman S, Shannon KM, Lange BJ. Childhood monosomy 7: epidemiology, biology, and mechanistic implications. *Blood* 85:1985–1999 (1995).
87. van den Berghe H, Vermaelen K, Mecucci C, Barbieri D,

- Tricot G. The 5q- anomaly. *Cancer Genet Cytogenet* 17:189-225 (1985).
88. Rowley JD, Le Beau MM. Cytogenetic and molecular analysis of therapy-related leukemia. *Ann NY Acad Sci* 567:130-140 (1989).
89. Murphree AL, Benedict WF. Retinoblastoma: clues to human oncogenesis. *Science* 223:1028-1033 (1984).
90. Nimer SD, Golde DW. The 5q- abnormality. *Blood* 70:1705-1712 (1987).
91. Ridge SA, Worwood M, Oscier D, Jacobs A, Padua RA. FMS mutations in myelodysplastic, leukemic, and normal subjects. *Proc Natl Acad Sci USA* 87:1377-1380 (1990).
92. Radich JP, Kopecky KJ, Appelbaum F, Willman CL, Collins SJ. N-ras mutations in acute myelogenous leukemia: a review of the current literature and an update of the Southwest Oncology Group experience. *Leuk Lymphoma* 6:325-334 (1992).
93. Liu ET. The role of ras gene mutations in myeloproliferative disorders. *Clin Lab Med* 10:797-807 (1990).
94. Kastan MB, Strauss LC, Civin CI. The role of hematopoietic growth factors and oncogenes in leukemogenesis. *Am J Pediatr Hematol Oncol* 11:249-267 (1989).
95. Needleman SW. Ras protooncogene activation in acute myeloid leukemia and related disorders. *Leuk Lymphoma* 5:85-91 (1991).
96. Taylor JA, Sandler DP, Bloomfield CD, Shore DL, Ball ED, Neubauer A, McIntyre OR, Liu E. Ras oncogene activation and occupational exposures in acute myeloid leukemia. *J Natl Cancer Inst* 84:1626-1632 (1992).
97. Kubo K, Naoe T, Kiyoi H, Fukutani H, Kato Y, Oguri T, Yamamori S, Akatsuka Y, Koda Y, Ohno R. Clonal analysis of multiple point mutations in the N-ras gene in patients with acute myeloid leukemia. *Jpn J Cancer Res* 84:379-387 (1993).
98. Hirai H, Okada M, Mizoguchi H, Mano H, Kobayashi Y, Nishida J, Takaku F. Relationship between an activated N-ras oncogene and chromosomal abnormality during leukemic progression from myelodysplastic syndrome. *Blood* 71:256-258 (1988).
99. Toksoz D, Farr CJ, Marshall CJ. Ras gene activation in a minor proportion of the blast population in acute myeloid leukemia. *Oncogene* 1:409-413 (1987).
100. Farr CJ, Saiki RK, Erlich HA, McCormick F, Marshall CJ. Analysis of RAS gene mutations in acute myeloid leukemia by polymerase chain reaction and oligonucleotide probes. *Proc Natl Acad Sci USA* 85:1629-1633 (1988).
101. Chen C-Y, Oliner JD, Zhan Q, Fornace AJ Jr, Vogelstein B, Kastan MB. Interactions between p53 and MDM2 in a mammalian cell cycle checkpoint pathway. *Proc Natl Acad Sci USA* 91:2684-2688 (1994).
102. Hollstein M, Sidransky D, Vogelstein B, Harris CC. p53 Mutations in human cancers. *Science* 253:49-53 (1991).
103. Soussi T, Jonveaux P. p53 Gene alterations in human hematological malignancies: a review. *Nouv Rev Fr Hematol* 33:477-480 (1991).
104. Knudson AG. Hereditary cancers disclose a class of cancer genes. *Cancer* 63:1888-1891 (1989).
105. Weinberg RA. The retinoblastoma protein and cell cycle control. *Cell* 81:323-330 (1995).
106. Kornblau S, Xu H-J, del Giglio A, Hu S-X, Zhang W, Calvert L, Beran M, Estey E, Andreeff M, Trujillo J, Cork MA, Smith TL, Benedict WF, Deisseroth AB. Clinical implications of decreased retinoblastoma protein expression in acute myelogenous leukemia. *Cancer Res* 52:4587-4590 (1992).
107. Furukawa Y, DeCaprio JA, Belvin M, Griffin JD. Heterogeneous expression of the product of the retinoblastoma susceptibility gene in primary human leukemia cells. *Oncogene* 6:1343-1346 (1991).
108. Bagby GC, Wilkinson B, McCall E, Lee M. Abnormalities of the hematopoietic regulatory network. *Adv Exp Med Biol* 241:255-264 (1988).
109. Foon KA, Champlin RE, Gale RP. Acute myelogenous leukemia and the myelodysplastic syndromes. In: *Cancer Treatment* (Haskell CM, ed). Philadelphia:W.B. Saunders, 1990; 589-606.
110. Holden JT, Geller RB, Farhi DC, Holland HK, Stempora LL, Phillips CN, Bray RA. Characterization of Thy-1 (CDw90) expression in CD34+ acute leukemia. *Blood* 86:60-65 (1995).
111. Sawada K, Sato N, Notoya A, Tarumi T, Hirayama S, Takano H, Koizumi K, Yasukouchi T, Yamaguchi M, Koike T. Proliferation and differentiation of myelodysplastic CD34+ cells: phenotypic subpopulations of marrow CD34+ cells. *Blood* 85:194-202 (1995).
112. Kroef MJPL, Fibbe WE, Mout R, Jansen RPM, Haak HL, Wessels JW, Van Kamp H, Willemze R, Landegent JE. Myeloid but not lymphoid cells carry the 5q deletion: polymerase chain reaction analysis of loss of heterozygosity using mini-repeat sequences on highly purified cell fractions. *Blood* 81:1849-1854 (1993).
113. Young DC, Wagner K, Griffin JD. Constitutive expression of the granulocyte-macrophage colony-stimulating factor gene in acute myeloblastic leukemia. *J Clin Invest* 79:100-106 (1987).
114. Zwierzina H, Schöllenberger S, Herold M, Schmalz J, Besemer J. Endogenous serum levels and surface receptor expression of GM-CSF and IL-3 in patients with myelodysplastic syndromes. *Leuk Res* 16:1181-1186 (1992).
115. Coutinho LH, Geary CG, Chang J, Harrison C, Testa NG. Functional studies of bone marrow haemopoietic and stromal cells in the myelodysplastic syndrome (MDS). *Br J Haematol* 75:16-25 (1990).
116. Greenberg BR, Wilson FD, Woo L. Granulopoietic effects of human bone marrow fibroblastic cells and abnormalities in the "granulopoietic microenvironment." *Blood* 58:557-564 (1981).
117. Sachs L, Lotem J. Control of programmed cell death in normal and leukemic cells: new implications for therapy. *Blood* 82:15-21 (1993).
118. Raza A, Mundle S, Iftikhar A, Gregory S, Marcus B, Khan Z, Alvi S, Shetty V, Dameron S, Wright V, Adler S, Loew JM, Shott S, Ali SN, Preisler H. Simultaneous assessment of cell kinetics and programmed cell death in bone marrow biopsies of myelodysplasias reveals extensive apoptosis as the probable basis for ineffective hematopoiesis. *Am J Hematol* 48:143-154 (1995).
119. Raza A, Gezer S, Mundle S, Gao X-Z, Alvi S, Borok R, Rifkin S, Iftikhar A, Shetty V, Parcharidou A, Loew J, Marcus B, Khan Z, Chaney C, Showel J, Gregory S, Preisler H. Apoptosis in bone marrow biopsy samples involving stromal and hematopoietic cells in 50 patients with myelodysplastic syndromes. *Blood* 86:268-276 (1995).
120. Pech N, Hermine O, Goldwasser E. Further study of internal autocrine regulation of multipotent hematopoietic cells. *Blood* 82:1502-1506 (1993).
121. Pedersen-Bjergaard J, Philip P, Larsen SO, Jensen G, Byrting K. Chromosome aberrations and prognostic factors in therapy-related myelodysplasia and acute nonlymphocytic leukemia. *Blood* 76:1083-1091 (1990).
122. Wald N, Conner MK. Induced chromosome damage after irradiation and cytotoxic drugs. *Hematology* 8:159-201 (1988).