REVIEW ARTICLE

MOLECULAR ORIGINS OF CANCER

Oncogenes and Cancer

Carlo M. Croce, M.D.

From the Department of Molecular Virology, Immunology, and Medical Genetics and the Human Cancer Genetics Program, Ohio State University Medical Center, Columbus. Address reprint requests to Dr. Croce at the Human Cancer Genetics Program and Department of Molecular Virology, Immunology, and Medical Genetics, Ohio State University Medical Center, 385L Wiseman Hall, 400 W. 12th Ave., Columbus, OH 43210, or at carlo.croce@osumc.edu.

N Engl J Med 2008;358:502-11.

Cancer is caused by alterations in oncogenes, tumor-suppressor genes, and microRNA genes. These alterations are usually somatic events, although germ-line mutations can predispose a person to heritable or familial cancer. A single genetic change is rarely sufficient for the development of a malignant tumor. Most evidence points to a multistep process of sequential alterations in several, often many, oncogenes, tumor-suppressor genes, or microRNA genes in cancer cells.

Tumors often possess cytogenetically different clones that arise from the initial transformed cell through secondary or tertiary genetic alterations. This heterogeneity contributes to differences in clinical behavior and responses to treatment of tumors of the same diagnostic type. Apart from the initial clone and subclones, tumors can also contain progenitor cancer cells, all of which constitute a spectrum of cells with different genetic alterations and states of differentiation. These populations can differ in sensitivity to chemotherapy, radiotherapy, and other treatments, making clinical management difficult. For these reasons, the initiating steps in the development of cancer are of considerable clinical importance and are a priority in the development of rational cancer treatment.

An example of this concept is chronic myelogenous leukemia, which is initiated by a reciprocal t(9;22) chromosomal translocation that fuses the *ABL* proto-oncogene to the *BCR* gene.[1,2] The fusion gene encodes an oncogenic ABL fusion protein with enhanced tyrosine kinase activity. All leukemic cells carry this chromosomal alteration, which is why inhibition of the excessive tyrosine kinase activity of the fusion protein by imatinib induces complete remission in most patients[3,4]; when relapse occurs, the leukemic cells usually carry mutations in *ABL* that render them resistant to the drug.[5]

EVIDENCE OF SOMATIC GENETIC CHANGE

The first evidence that cancer arises from somatic genetic alterations came from studies of Burkitt's lymphoma, in which one of three different translocations juxtaposes an oncogene, *MYC*, on chromosome 8q24 to one of the loci for immunoglobulin genes. Chromosomes 14q, 22q, and 2p — the translocation partners — each carries enhancer elements in the immunoglobulin loci, thereby activating the juxtaposed *MYC* oncogene (see Fig. 1 in the Supplementary Appendix, available with the full text of this article at www.nejm.org).[6-11] Since every malignant lymphocyte carries the *MYC* translocation, deregulation of the *MYC* oncogene is probably the initiating event.

Second, transfection experiments have shown that mouse fibroblasts, when transfected in vitro with DNA from human cancer cells, acquire some of the properties of malignant cells (i.e., transformation). The transforming activity of the DNA was traced to a human homologue of the retroviral *RAS* oncogene. This oncogene bears mutations that activate the transforming property of the RAS oncogenic protein.[12,13]

Third, the cloning and characterization of the chromosomal breakpoints that are characteristic of follicular lymphomas and some diffuse large B-cell lymphomas[14] have shown a juxtaposition of the *BCL2* oncogene to enhancer elements in the immunoglobulin heavy-chain locus, resulting in deregulation of *BCL2*[14,15] (see Fig. 2 in the Supplementary Appendix).

Fourth, in transgenic mice that carry an activated oncogene from a human tumor, cancers develop that resemble the human tumor.[16,17] That these cancers appear only after a latent period suggests that alterations in other genes must occur before progression to frank neoplasia can occur — activation of a particular oncogene seems to be necessary but not sufficient for the development of overt cancer.

FUNCTIONAL PROPERTIES OF ONCOGENES

Historically, transformation events in cancer have been defined as initiation events (contributing to the early stages of neoplastic transition) or progression events (referring to subsequent transformative processes). Oncogenes encode proteins that control cell proliferation, apoptosis, or both. They can be activated by structural alterations resulting from mutation or gene fusion,[18] by juxtaposition to enhancer elements,[19] or by amplification. Translocations and mutations can occur as initiating events[20] or during tumor progression, whereas amplification usually occurs during progression. (Table 1 in the Supplementary Appendix lists oncogenes in tumors of different species, the methods used to identify them, their mechanisms of activation, and the functions of their encoded products; Table 2 in the Supplementary Appendix lists the molecularly characterized chromosomal rearrangements in human cancers.) The products of oncogenes can be classified into six broad groups: transcription factors, chromatin remodelers, growth factors, growth factor receptors, signal transducers, and apoptosis regulators.

PRODUCTS OF ONCOGENES

Transcription Factors

Transcription factors are often members of multigene families that share common structural domains. To act, many transcription factors require interaction with other proteins. In some tumors, for example, the Fos transcription protein dimerizes with the Jun transcription factor to form the AP1 transcription factor, and this complex increases the expression of several genes that control cell division.[21,22]

Chromosomal translocations often activate transcription-factor genes in lymphoid cancers[23] and sometimes do so in solid tumors (e.g., prostate cancer[24]; see Table 2 in the Supplementary Appendix). In certain sarcomas, chromosomal translocations that result in fused proteins occur consistently; in Ewing's sarcoma, for example, the *EWS* gene is fused with one of a number of partner genes, resulting in aberrant transcriptional activity of the fused proteins (see Table 2 in the Supplementary Appendix). The EWS protein is an RNA-binding molecule with a domain that, when fused to a heterologous DNA-binding domain, can greatly stimulate gene transcription. Prostate carcinomas carry translocations of the *TMPR552* gene that fuse with and activate *ERG1* or *ETV1.* These genes are members of the ETS family of transcription regulators, which can activate or repress genes involved in cellular proliferation, differentiation, and apoptosis. The fusion of *TMPR552,* which has androgen-responsive promoter elements, with an ETS-related gene creates a fusion protein that increases proliferation and inhibits apoptosis of cells in the prostate gland, thereby facilitating their transformation into cancer cells.[24]

Chromatin Remodelers

Modifications in the degree of compaction of chromatin play a critical role in the control of gene expression, replication, and repair and of chromosome segregation. Two kinds of enzymes remodel chromatin: ATP-dependent enzymes[25] that move the positions of nucleosomes, the repeating subunits of the histones in chromatin around which DNA winds, and enzymes that modify the N-terminal tails of histones.[26] The pattern of histone modification constitutes an epigenetic code that determines the interaction between nucleosomes and chromatin-associated proteins.[27] These interactions, in turn, determine the structure of chromatin and its transcriptional capacity.

In acute lymphocytic leukemia and acute myelogenous leukemia, the *ALL1* (also named *MLL*) gene can fuse with 1 of more than 50 genes. ALL1 is part of a very large, stable multiprotein complex. Most of the proteins in the complex are

components of transcription complexes[28]; others are involved in histone methylation and RNA processing. The entire complex remodels, acetylates, deacetylates, and methylates nucleosomes and free histones.[28] The fusion of ALL1 with 1 of more than 50 proteins results in the formation of the chimeric proteins that underlie acute lymphoblastic leukemia and acute myelogenous leukemia. ALL1 (MLL) fusion proteins deregulate homeobox genes (which encode transcriptions factors), the *EPHA7* gene (which encodes a receptor tyrosine kinase), and microRNA genes such as *miR191.*

Growth Factors

Constitutive activation of a growth factor gene can contribute to malignant transformation. Platelet-derived growth factor (PDGF) consists of α and β chains and is released from platelets during coagulation.[29] It can induce the proliferation of various cell types and stimulate fibroblasts to participate in wound healing. The *sis* oncogene of simian sarcoma virus is structurally similar to the gene for the β chain of PDGF.[29] Overexpression of PDGF induces the in vitro transformation of fibroblasts containing PDGF receptors; it does not influence fibroblasts lacking these receptors. This autocrine loop entails overexpression of *PDGF-β*, the expression of the PDGF-β receptor, and unregulated cell growth. An antibody against PDGF-β, an antibody against its receptor, or small molecules that block the receptor inhibit growth of the transformed fibroblasts.

The WNT family of secreted glycoproteins inhibits phosphorylation of β-catenin, which is involved in cell–cell adhesion and the activation of several signal-transduction pathways[30] (Fig. 1). The APC protein controls the activity of β-catenin. In familial adenomatous polyposis, inactivating mutations of *APC* block the degradation of β-catenin by inhibiting its phosphorylation. As a result, free β-catenin in the cytoplasm

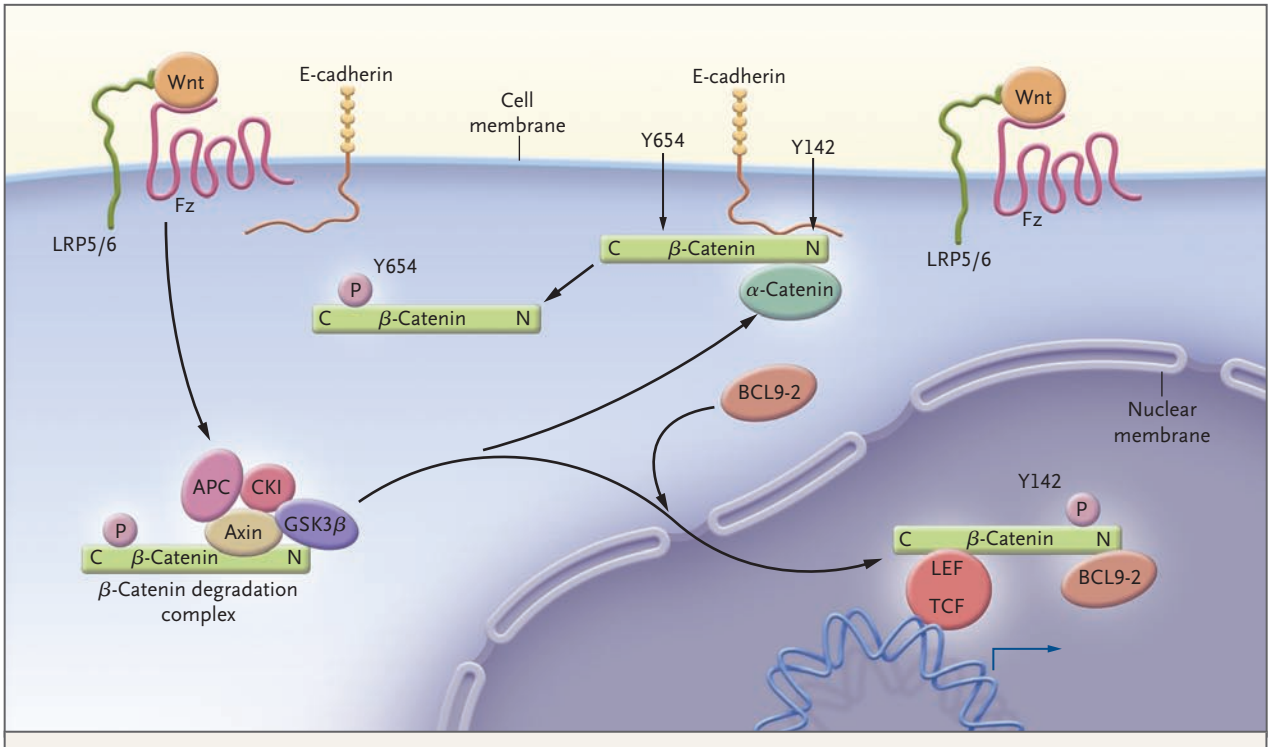

Figure 1. Dual Functions of β-Catenin in Cell Adhesion and Transcription.

β-Catenin is in a destructive cytoplasmic complex composed of activated protein C (APC), axin, glycogen synthase kinase 3 beta (GSK3β), and casein kinase (CK1). CK1 and GSK3β induce serine–threonine phosphorylation of the N-terminal of β-catenin. Binding of Wnt ligands to the frizzled (F_z), LRP5, and LRP6 receptors inhibits the degradation of this complex and leads to nuclear accumulation of β-catenin. Phosphorylation (P) of tyrosine 142 of β-catenin leads it to interact with BCL9-2 and to migrate to the nucleus, where the β-catenin–BCL9-2 complex binds LEF and TCF to induce the expression of target genes. Phosphorylation of tyrosine Y654 of β-catenin results in the disassociation of E-cadherin and β-catenin, causing loss of cell–cell adhesion and increased cell motility.[6-11]

translocates to the nucleus, where it activates genes involved in cell proliferation and invasion (Fig. 1).

Growth Factor Receptors

Growth factor receptors are altered in many cancers (Fig. 2).[31] In many tumors, a deletion of the ligand-binding domain of epidermal growth factor receptor (EGFR), a transmembrane protein with tyrosine kinase activity, causes constitutive activation of the receptor in the absence of ligand binding.[32] The activated receptor phosphorylates tyrosines in the intracellular domain of the receptor, providing interaction sites for cytoplasmic proteins containing the SRC homology domain and other binding domains. These interactions deregulate signaling in several pathways. Activating mutations occur in three other members of the EGFR family — ERBB2, ERBB3, and ERBB4 — and within the kinase domains of the HER2/neu and KIT signaling receptors. Such mutations occur in lung and breast cancer and gastrointestinal stromal tumors. Two classes of clinically active anti-EGFR agents have been developed: a monoclonal antibody against the extracellular domain of the receptor (cetuximab) and competitive inhibitors of the tyrosine kinase activity of the receptor (e.g., erlotinib and gefitinib).

Vascular endothelial growth factor (VEGF) regulates hypoxia-dependent control of gene transcription (Fig. 3). The activity of VEGF is mediated by three receptor tyrosine kinases: VEGFR1 (FLT1), VEGFR2 (FLK1-KDR), and VEGFR3 (FLT4). VEGF stimulates angiogenesis in a variety of cancers, and inhibitors of VEGF and of the VEGFRs have been developed. Bevacizumab is a monoclonal anti-VEGF antibody, and SU5412, a small molecule, binds the receptor tyrosine kinases of VEGFR1 and VEGFR2 as well as the kinases of the PDGF receptor and KIT. In addition to inhibiting the ABL kinase, imatinib also inhibits the PDGF and KIT receptor kinases. Gastrointestinal stromal tumors that carry activating mutations of *KIT*[33,34] respond to imatinib or other inhibitors of these receptor kinases.

Signal Transducers

Binding of receptor tyrosine kinases to the appropriate ligand causes reorganization of the receptors and autophosphorylation of tyrosines in the intracellular portion of the molecules[35] (Fig. 2). Autophosphorylation enhances the kinase ac-

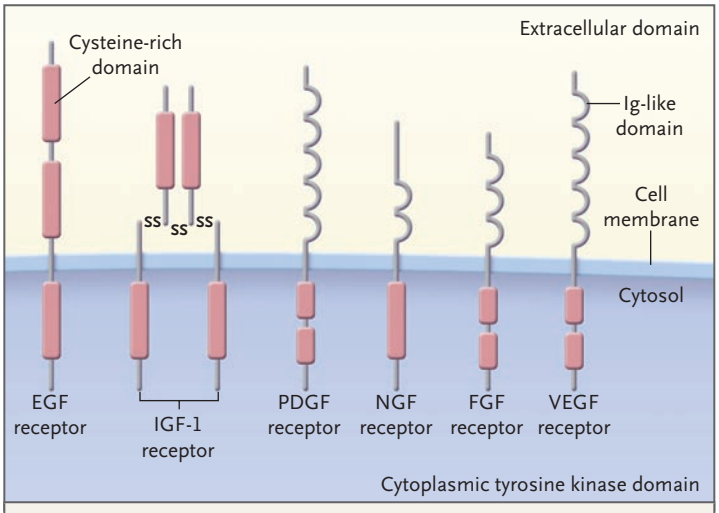

Figure 2. Examples of Receptor Tyrosine Kinases.

The epidermal growth factor (EGF), insulin-like growth factor 1 (IGF-1), platelet-derived growth factor (PDGF), and fibroblast growth factor (FGF) receptors have been found to be involved in a variety of human cancers. NGF denotes nerve growth factor, SS disulfide bonds, and VEGF vascular endothelial growth factor.

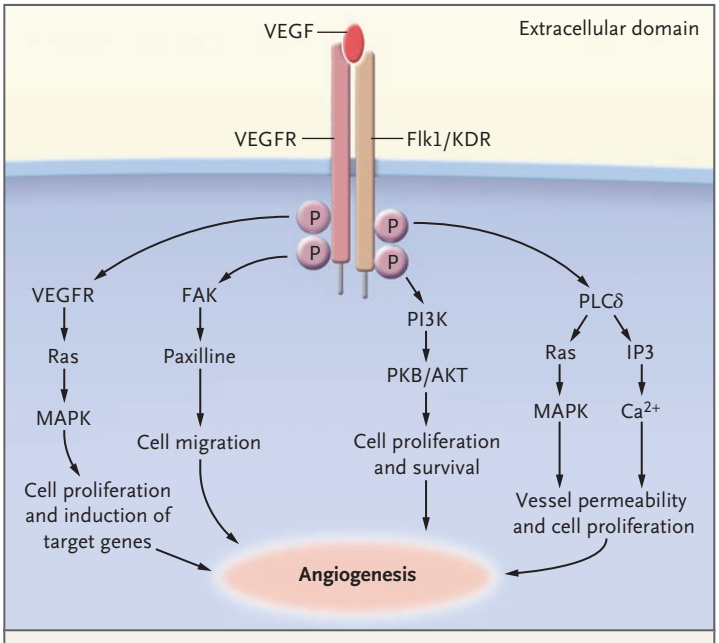

Figure 3. Role of VEGF–VEGFR Interaction in Angiogenesis.

Several pathways are activated by the interaction of vascular endothelial growth factor (VEGF) and VEGF receptors (VEGFR). FAK denotes fatal adhesion kinase, Flk fetal liver kinase, IP3 inositol triphosphate, KDR kinase-insert domain–containing receptor, MAPK mitogen-activated protein kinase, PI3K phosphoinositol 3-kinase, PKB protein kinase B, and PLC phospholipase C.

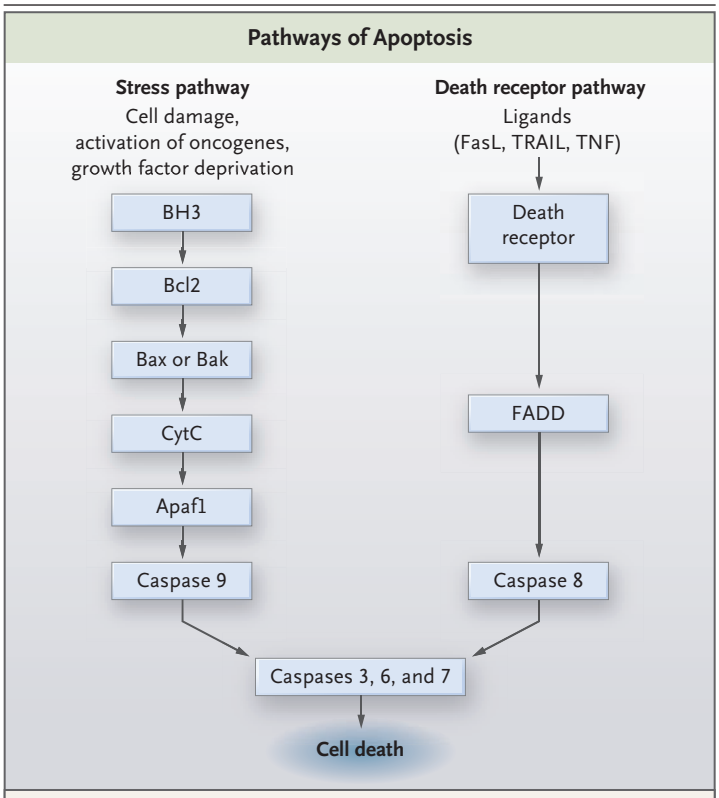

Figure 4. The Two Main Pathways to Programmed Cell Death, or Apoptosis.
The effectors of cell death are the downstream caspases, proteolytic enzymes activated by caspases 8 and 9, which are capable of clearing many of the cellular proteins causing cell death. FADD denotes Fas-associated death domain.

tivity of the receptor or promotes the interaction of the receptor with domains of cytoplasmic proteins (e.g., the SRC homology 2 domain) that are effectors and regulators of intracellular signaling.[36] In humans, there are approximately 120 SRC homology 2 domains in 100 different proteins that mediate responses to signals initiated by phosphorylated tyrosines. Some of these proteins share domains with enzymatic activity, whereas others link activated receptors to downstream targets.

Many oncogenes encode members of signal-transduction pathways. They fall into two main groups: nonreceptor protein kinases and guanosine-triphosphate–binding proteins.[37,38] The nonreceptor protein kinases are of two types: tyrosine kinases (e.g., ABL, LCK, and SRC) and serine and threonine kinases (e.g., AKT, RAF1, MOS, and PIM1). Proteins involved in signal transduction become oncogenic if they bear activating mutations. An important example is PI3K and some of its downstream targets, such as AKT and SGK, which are critical to tyrosine kinase signaling and can be mutated in cancer cells.

Apoptosis Regulators

The *BCL2* gene, which is involved in the initiation of almost all follicular lymphomas and some diffuse large B-cell lymphomas (see Fig. 2 in the Supplementary Appendix),[14,15] encodes a cytoplasmic protein[39,40] that localizes to mitochondria and increases cell survival by inhibiting apoptosis.[41] *BCL2* is also important in chronic lymphocytic leukemia and lung cancer. The BCL2 family members BCL-XL and BCL2 inhibit apoptosis and are up-regulated in many cancers.

Two main pathways lead to apoptosis: the stress pathway and the death-receptor pathway (Fig. 4). The stress pathway is triggered by proteins that contain the BCL2 homology 3 domain; this domain inactivates BCL2 and BCL-XL (which normally inhibit apoptosis) and thereby activates the caspases that induce apoptosis (Fig. 4). Drugs that mimic the BCL2 homology 3 domain and can bind to BCL-XL or BCL2 (peptides or small organic molecules that bind in a groove of these proteins) are under development. This approach has attracted considerable attention because many tumors overexpress BCL2 or related proteins. The death-receptor pathway is activated by the binding of Fas ligand, TRAIL, and tumor necrosis factor α, to their corresponding (death) receptors on the cell surface. Activation of death receptors activates caspases that cause cell death (Fig. 4).

ONCOGENE ACTIVATION

Activation of oncogenes by chromosomal rearrangements, mutations, and gene amplification confers a growth advantage or increased survival of cells carrying such alterations. All three mechanisms cause either an alteration in the oncogene structure or an increase in or deregulation of its expression.[42]

Chromosomal Rearrangements

Chromosome inversions and translocations are common cytogenetic abnormalities in cancer cells. In hematopoietic cancers and solid tumors, the translocations and inversions increase or deregulate transcription of the oncogene. In prostate cancer, gene fusion occurs between a gene that carries a promoter that is very active in the target

cells, and another that carries the oncogenic activity (e.g., *ERG1*).[24] In cancers of B and T cells, the most common mechanism of activation by translocation resembles *MYC* deregulation, whereas in myeloid cancers and soft-tissue sarcomas, gene fusion is more common (see Table 2 in the Supplementary Appendix).

Mutations

When an oncogene is activated by mutation, the structure of the encoded protein is changed in a way that enhances its transforming activity. Many types of mutation occur in oncogenes.[43] Examples are the *RAS* oncogenes (*KRAS, HRAS,* and *NRAS*), which encode proteins with guanosine-nucleotide–binding activity and intrinsic guanosine triphosphatase activity. When mutated in codon 12, 13, or 61, the *RAS* genes encode a protein that remains in the active state and continuously transduces signals by linking tyrosine kinases to downstream serine and threonine kinases. These incessant signals induce continuous cell growth. Mutation of oncogenes in the RAS family has been associated with exposure to environmental carcinogens. Mutations of *KRAS* are common in carcinomas of the lung, colon, and pancreas,[43] whereas mutations of *NRAS* occur principally in acute myelogenous leukemia and the myelodysplastic syndrome.[44]

Activating point mutations of the *BRAF* gene occur in 59% of melanomas, 18% of colorectal cancers, 14% of hepatocellular carcinomas, and 11% of gliomas.[45] Most of the *BRAF* mutations change the valine residue at position 599 to glutamic acid (V599E). This change occurs within the kinase domain of the BRAF protein, resulting in a constitutively active protein that uncontrollably stimulates the MAP kinase cascade, thereby deregulating genes involved in cell proliferation, differentiation, and survival.[45,46] In melanoma, *BRAF* mutations can precede neoplastic transformation; several types of nevi carry *BRAF* mutations.

Gene Amplification

An example of gene amplification, which usually occurs during tumor progression, is the amplification of the dihydrofolate reductase gene (*DHFR*) in methotrexate-resistant acute lymphoblastic leukemia.[47] Amplification of *DHFR* is accompanied by cytogenetic alterations that mirror amplification of oncogenes.[48,49] The amplified DNA segment usually involves several hundred kilobases and can contain many genes. Members of four different oncogene families are often amplified: MYC, cyclin D1 (or CCND1), EGFR, and RAS. *MYC* is amplified in small-cell lung cancer, breast cancer, esophageal cancer, cervical cancer, ovarian cancer, and head and neck cancer, whereas amplification of *NMYC* correlates with an advanced tumor stage.[50] The t(11;14) translocation juxtaposes *CCND1* and immunoglobulin enhancer elements and is characteristic of mantle-cell lymphoma.[14] *CCND1* amplification also occurs in breast, esophageal, hepatocellular, and head and neck cancer. *EGFR* (*ERBB1*) is amplified in glioblastoma and head and neck cancer. Amplification of *ERBB2* (also called *HER2/neu*) in breast cancer correlates with a poor prognosis.[51] A monoclonal antibody against the product of this oncogene (trastuzumab) is effective in breast cancers that overexpress HER2/neu. Table 3 in the Supplementary Appendix lists oncogenes that are amplified in different types of cancer.

ONCOGENES IN CANCER INITIATION AND PROGRESSION

When chronic myelogenous leukemia converts to acute leukemia, the malignant clone acquires an additional t(9;22) translocation, an isochromosome 17, or trisomy of chromosome 8. When follicular lymphoma becomes aggressive, the lymphoma cells often bear a t(8;14) translocation in addition to the original t(14;18) translocation. These findings support the hypothesis that most hematopoietic tumors and soft-tissue sarcomas are initiated by the activation of an oncogene, followed by alterations in tumor-suppressor genes and other oncogenes. In contrast, most carcinomas are initiated by the loss of function of a tumor-suppressor gene, followed by alterations in oncogenes and additional tumor-suppressor genes.[52,53] This multistep process in human cancer has also been found in mouse models carrying activated oncogenes or inactivated tumor-suppressor genes, in which the duration and aggressiveness of the disease can be changed by introducing into the mouse genome the same sequential genetic alterations observed in human tumors. Methylation of CpG islands located in the promoter regions of a number of tumor-suppressor genes has also been considered an important epigenetic step in the process of carcinogenesis. This topic will be covered later in this series.

Table 1. Cancer Therapies That Target Oncogenic Proteins.*

Anticancer Drug	Target	Disease
Monoclonal antibodies		
Trastuzumab (Herceptin, Genentech)	ERBB2	Breast cancer
Cetuximab (Erbitux, ImClone)	EGFR	Colorectal cancer
Bevacizumab (Avastin, Genentech)	VEGF	Colorectal cancer, non–small-cell lung cancer
Small molecules		
Imatinib (Gleevec, Novartis)	ABL, PDGFR, KIT	Chronic myelogenous leukemia, gastrointestinal stromal tumors, chordoma
Gefitinib (Iressa, AstraZeneca)	EGFR	Non–small-cell lung cancer
Erlotinib (Tarceva, Genentech)	EGFR	Non–small-cell lung cancer
Sorafenib (Nexavar, Bayer/Onyx)	VEGFR, PDGFR, FLT3	Renal-cell carcinoma
Sunitinib (Sutent, Pfizer)	VEGFR, PDGFR, FLT3	Gastrointestinal stromal tumors, renal-cell carcinoma

* EGFR denotes epidermal growth factor receptor, FLT3 FMS-like tyrosine kinase 3, PDGFR platelet-derived growth factor receptor, and VEGF vascular endothelial growth factor.

ONCOGENES AS THERAPEUTIC TARGETS

Oncogenic proteins in cancer cells can be targeted by small molecules and, when the oncogenic protein is expressed on the cell surface, by monoclonal antibodies. Table 1 contains a summary of the targets and drugs (small molecules and monoclonal antibodies) being used in the treatment of a variety of human cancers.

Imatinib targets the initial step of the multistep process in chronic myelogenous leukemia.[54] The same drug can affect the KIT and PDGFR receptor kinases.[55,56] Of particular interest are inhibitors of the BCL2 family, which can induce the apoptotic death of cancer cells. In acute promyelocytic leukemia, which is initiated by a t(15;17) chromosome translocation that fuses the *PML* gene to *RARα* (a nuclear receptor for retinoic acids[57-59]; see Table 2 in the Supplementary Appendix), retinoic acid can induce terminal differentiation and death of APL cells. This modality is called differentiation therapy.

MicroRNA GENES

MicroRNA genes, unlike other genes involved in cancer, do not encode proteins. Instead, the products of these genes consist of a single RNA strand of about 21 to 23 nucleotides; their function is to regulate gene expression. A microRNA molecule can anneal to a messenger RNA (mRNA) containing a nucleotide sequence that complements the sequence of the microRNA (Fig. 5). In this way, the microRNA blocks protein translation or causes degradation of the mRNA. Examples of the role microRNA plays in cancer pathophysiology involve *miR-15a* and *miR-16-1*, which are deleted or down-regulated in most indolent cases of chronic lymphocytic leukemia, suggesting an early event in the pathogenesis of this disease.[59]

Mapping of numerous microRNA genes has shown that many occur in chromosomal regions that undergo rearrangements, deletions, and amplifications in cancer cells.[60] The regions of the genome that are consistently involved in chromosomal rearrangements in cancer cells but that lack oncogenes or tumor-suppressor genes appear to harbor microRNA genes.

Expression profiling of microRNA genes has revealed signatures associated with tumor classification, diagnosis, staging, and progression, as well as prognosis and response to treatment.[61-63] For example, microRNA expression profiling can distinguish between indolent and aggressive forms of chronic lymphocytic leukemia,[62] and expression of a small panel of microRNA genes correlates with prognosis in stage 1 lung cancer.[63] Some microRNA genes that are deregulated in chronic lymphocytic leukemia have germ-line or somatic mutations in a microRNA precursor that affect the processing of short single-stranded microRNA molecules.[62] MicroRNA genes can be up-regulated or down-regulated in cancer cells.[64] The up-regulated genes function as oncogenes by down-regulating tumor-suppressor genes, whereas the down-regulated genes function as tumor-

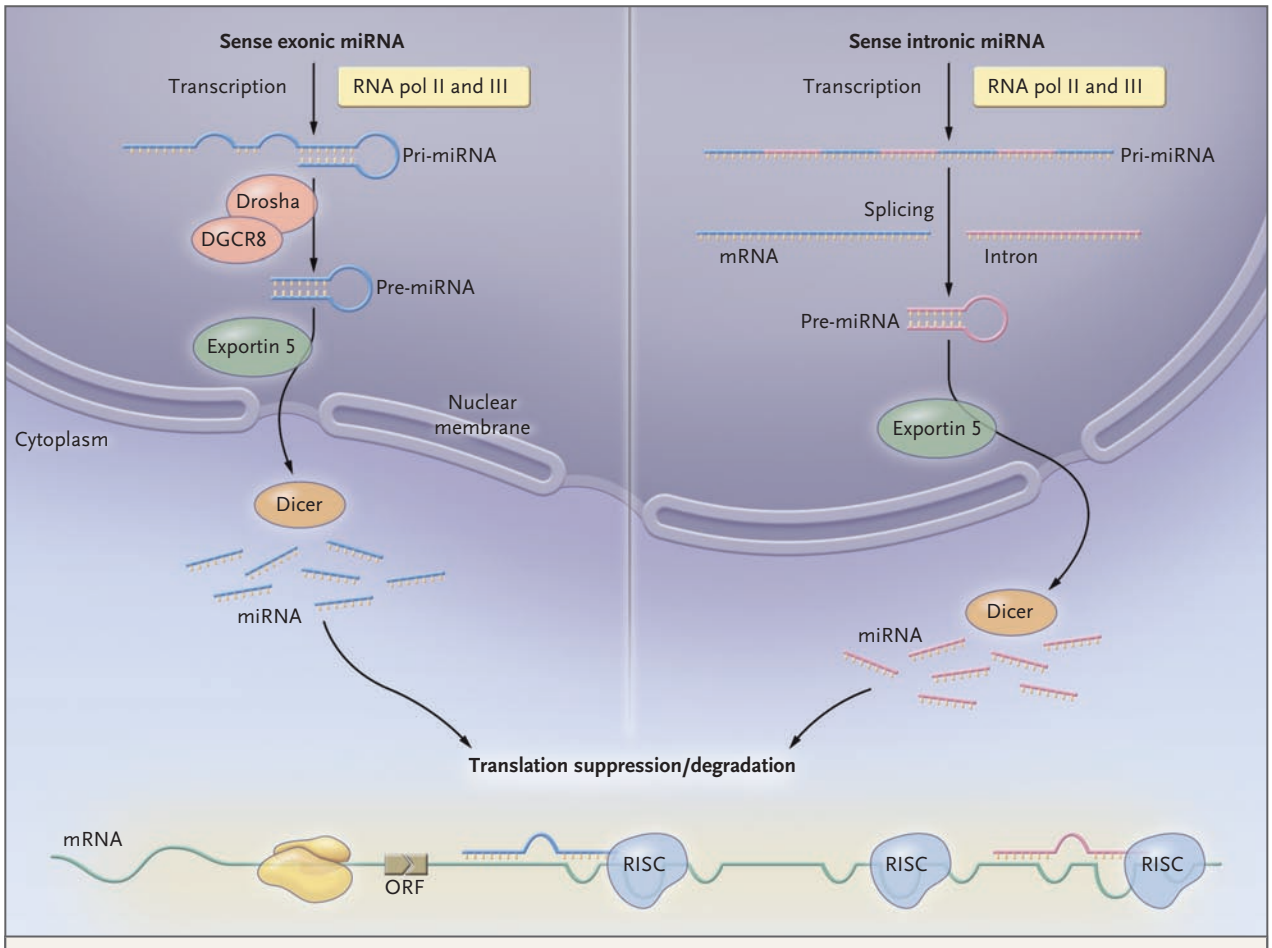

Figure 5. Mechanisms Involved in the Expression of Mature Exonic and Intronic MicroRNAs.

MicroRNA (miRNA) is transcribed mostly by RNA polymerase (pol) II and less frequently by RNA pol III. The primary transcript (pri-microRNA) can be quite large. During the process of splicing, pri-microRNA is processed in the nucleus by an enzymatic complex that includes Drosha and DGCR8, which leads to the formation of a smaller (70-to-100 nucleotide), second hairpin precursor named pre-microRNA. This second precursor binds exportin-5 in the nucleus and is transported to the cytoplasm, where it is cleaved by Dicer into mature microRNA. This mature microRNA, for the most part, binds to the 3' untranslated region of messenger RNA (mRNA) and, depending on the degree of complementarity with the target RNA, can lead to the degradation or blockage of translation mRNA. Recent studies suggest that translation blockage is accompanied by some degradation.

suppressor genes by down-regulating oncogenes. The function of microRNA genes depends on their targets in a specific tissue. A microRNA gene can be a tumor suppressor if in a given cell type its critical target is an oncogene, and it can be an oncogene if in a different cell type its target is a tumor-suppressor gene.

Up-regulation of microRNA genes can be due to amplification, deregulation of a transcription factor, or demethylation of CpG islands in the promoter regions of the gene. For example, the ALL1 (MLL) fusion proteins of acute lymphoblastic leukemia or acute myeloblastic leukemia carrying chromosome 11q23 translocations target the Drosha nuclease complex to specific microRNA genes, including *miR191*, thereby enhancing the processing of their microRNA precursors.[65] The *miR191* gene is also up-regulated in numerous types of solid cancers,[64] suggesting that it is the downstream target of signal-translocation pathways involved in cancer.[65] MicroRNA genes functioning as tumor suppressors can be down-regulated because of deletions, epigenetic silencing, or loss of the expression of one or more transcription factors.

The *miR155* gene is overexpressed in diffuse large B-cell lymphoma,[66] the aggressive form of chronic lymphocytic leukemia,[62] and in breast,

lung, and colon cancers. In transgenic mice carrying this gene under control of the Eμ enhancer of immunoglobulin genes,[67] overexpression of *miR155* causes acute lymphoblastic leukemia or high-grade lymphoma, indicating that deregulation of a single microRNA gene can cause malignant transformation.[67] Since it takes several months for the tumors in these mice to become aggressive, it is likely that additional genetic alterations are needed for the development of frank neoplasia.

Members of the LET7 microRNA family, which are deleted or underexpressed in lung cancer, target *RAS*[68]; loss of *LET7* results in overexpression of *RAS*.[68] *MiR15a* and *miR-16-1*, the microRNAs that are deleted or down-regulated in chronic lymphocytic leukemia, cause overexpression of *BCL2*, which protects cells from apoptosis (Fig. 4).[69] The expression of a set of 21 microRNAs is altered in at least three types of solid tumors.[64] One of these 21 genes, *miR21*, is of particular interest because it inhibits expression of the tumor suppressor PTEN.[70] PTEN encodes a phosphatase involved in the PI3K kinase signaling pathway and is deleted, mutated, or silenced in advanced breast, lung, gastric, and prostate cancers.[70]

SUMMARY

The identification of oncogenes involved in the initiation and progression of tumors has generated targets for the development of new anticancer drugs. Several new drugs, small molecules, and monoclonal antibodies directly affecting oncogene products have been developed, and more will follow. Considerable progress has been made in producing small molecules capable of inhibiting the enzymatic activity of ABL, KIT, EGFR, and ERBB2. For cases in which the oncogene products are not enzymes, it has been much more difficult to develop new agents.

The advantage of targeted therapy is the dependency of cancer cells on the oncogene product for growth and survival. Thus, cancer cells are more sensitive to the treatment than are normal cells. All targets, however, are not equivalent. It is possible to foresee the development of multiple drugs that have multiple targets involved in the development of cancer. The discovery of the involvement of microRNAs in the initiation and progression of human cancer may provide additional targets for anticancer treatments.

No potential conflict of interest relevant to this article was reported.

REFERENCES

1. Groffen J, Stephenson JR, Heisterkamp N, et al. Philadelphia chromosomal breakpoints are clustered within a limited region, bcr, on chromosome 22. Cell 1984;36:93-4.
2. Shtivelman E, Lifshitz B, Gale RP, Canaani E. Fused transcript of abl and bcr genes in chronic myelogenous leukemia. Nature 1985;315:550-4.
3. Druker BJ, Talpaz M, Resta DJ, et al. Efficacy and safety of a specific inhibitor of the BCR-ABL tyrosine kinase in chronic myeloid leukemia. N Engl J Med 2001;344:1031-7.
4. Ottmann OG, Druker BJ, Sawyers CL, et al. A phase 2 study of imatinib in patients with relapsed or refractory Philadelphia chromosome-positive acute lymphoid leukemias. Blood 2002;100:1965-71.
5. Roche-Lestienne C, Soenen-Cornu V, Grardel-Duflos N, et al. Several types of mutations of the Abl gene can be found in chronic myeloid leukemia patients resistant to STI571, and they can pre-exist to the onset of treatment. Blood 2002;100:1014-8.
6. Croce CM, Shander M, Martinis J, et al. Chromosomal location of the genes for human immunoglobulin heavy chain. Proc Natl Acad Sci U S A 1979;76:3416-9.
7. Erikson J, Martinis J, Croce CM. Assignment of the genes for human lambda immunoglobulin chains to chromosome 22. Nature 1981;294:173-5.
8. Dalla Favera R, Bregni M, Erikson J, Patterson D, Gallo RC, Croce CM. Human c-myc onc gene is located on the region of chromosome 8 that is translocated in Burkitt lymphoma cells. Proc Natl Acad Sci U S A 1982;79:7824-7.
9. Croce CM, Thierfelder W, Erikson J, et al. Transcriptional activation of an unrearranged and untranslocated c-myc oncogene by translocation of a C lambda locus in Burkitt lymphoma. Proc Natl Acad Sci U S A 1983;80:6922-6.
10. Erikson J, Nishikura K, ar-Rushdi A, et al. Translocation of an immunoglobulin kappa locus to a region 3′ of an unrearranged c-myc oncogene enhances c-myc transcription. Proc Natl Acad Sci U S A 1983;80:7581-5.
11. ar-Rushdi A, Nishikura K, Erikson J, Watt R, Rovera G, Croce CM. Differential expression of the translocated and the untranslocated c-myc oncogene in Burkitt lymphoma. Science 1983;222:390-3.
12. Capon DJ, Chen EY, Levinson AD, Seeburg PH, Goeddel DV. Complete nucleotide sequences of the T24 human bladder carcinoma oncogene and its normal homologue. Nature 1983;302:33-7.
13. McCoy MS, Toole JJ, Cunningham JM, Chang EH, Lowy DR, Weinberg RA. Characterization of a human colon/lung carcinoma oncogene. Nature 1983;302:79-81.
14. Tsujimoto Y, Yunis J, Onorato-Showe L, Nowell PC, Croce CM. Molecular cloning of the chromosomal breakpoint of B cell lymphomas and leukemias with the t(11;14) chromosome translocation. Science 1984;224:1403-6.
15. Tsujimoto Y, Cossman J, Jaffe E, Croce CM. Involvement of the bcl-2 gene in human follicular lymphoma. Science 1985;228:1440-3.
16. Adams JM, Harris AW, Pinkert CA, et al. The c-myc oncogene driven by immunoglobulin enhancers induces lymphoid malignancy in transgenic mice. Nature 1985;318:533-8.
17. Leder A, Pattengale PK, Kuo A, Stewart TA, Leder P. Consequences of widespread deregulation of the c-myc gene in transgenic mice: multiple neoplasms and normal development. Cell 1986;45:485-95.
18. Konopka JB, Watanabe SM, Singer JW, Collins SJ, Witte ON. Cell lines and clinical isolates derived from Ph1-positive chronic myelogenous leukemia patients express c-abl proteins with a common structural alteration. Proc Natl Acad Sci U S A 1985;82:1810-4.

19. Tsujimoto Y, Gorham J, Cossman J, Jaffe E, Croce CM. The t(14;18) chromosome translocations involved in B-cell neoplasms result from mistakes in VDJ joining. Science 1985;229:1390-3.
20. Finger LR, Harvey RC, Moore RCA, Showe LC, Croce CM. A common mechanism of chromosomal translocation in T- and B-cell neoplasia. Science 1986;234:982-5.
21. Shaulian E, Karin M. AP-1 in cell proliferation and survival. Oncogene 2001;20:2390-400.
22. *Idem.* AP-1 as a regulator of cell life and death. Nat Cell Biol 2002;4(5):E131-E136.
23. Croce CM. Role of chromosome translocations in human neoplasia. Cell 1987;49:155-6.
24. Tomlins SA, Rhodes DR, Perner S, et al. Recurrent fusion of TMPRSS2 and ETS transcription factor genes in prostate cancer. Science 2005;310:644-8.
25. Peterson CL, Workman JL. Promoter targeting and chromatin remodeling by the SWI/SNF complex. Curr Opin Genet Dev 2000;10:187-92.
26. Jenuwein T, Allis CD. Translating the histone code. Science 2001;293:1074-80.
27. Strahl BD, Allis CD. The language of covalent histone modifications. Nature 2000;403:41-5.
28. Nakamura T, Mori T, Tada S, et al. ALL-1 is a histone methyltransferase that assembles a supercomplex of proteins involved in transcriptional regulation. Mol Cell 2002;10:1119-28.
29. Heldin CH, Westermark B. Mechanism of action and in vivo role of platelet-derived growth factor. Physiol Rev 1999;79:1283-316.
30. Giles RH, van Es JH, Clevers H. Caught up in a Wnt storm: Wnt signaling in cancer. Biochim Biophys Acta 2003;1653:1-24.
31. Heldin CH. Dimerization of cell surface receptors in signal transduction. Cell 1995;80:213-23.
32. Arteaga CL. Epidermal growth factor receptor dependence in human tumors: more than just expression? Oncologist 2002;7:Suppl 4:31-9.
33. Joensuu H, Dimitrijevic S. Tyrosine kinase inhibitor imatinib (STI571) as an anticancer agent for solid tumours. Ann Med 2001;33:451-5.
34. Tamborini E, Bonadiman L, Greco A, et al. A new mutation in the KIT ATP pocket causes acquired resistance to imatinib in a gastrointestinal stromal tumor patient. Gastroenterology 2004;127:294-9.
35. Pawson T, Warner N. Oncogenic rewiring of cellular signaling pathways. Oncogene 2007;26:1268-75.
36. Sicheri F, Moarefi I, Kuriyan J. Crystal structure of the Src family tyrosine kinase Hck. Nature 1997;385:602-9.
37. Kaziro Y, Itoh H, Kozasa T, Nakafuku M, Satoh T. Structure and function of signal-transducing GTP-binding proteins. Annu Rev Biochem 1991;60:349-400.
38. Salgia R, Skarin AT. Molecular abnormalities in lung cancer. J Clin Oncol 1998;16:1207-17.
39. Tsujimoto Y, Croce CM. Analysis of the structure, transcripts, and protein products of bcl-2, the gene involved in human follicular lymphoma. Proc Natl Acad Sci U S A 1986;83:5214-8.
40. Tsujimoto Y, Ikegaki N, Croce CM. Characterization of the protein product of bcl-2, the gene involved in human follicular lymphoma. Oncogene 1987;2:3-7.
41. Adams JM, Cory S. The Bcl-2 apoptotic switch in cancer development and therapy. Oncogene 2007;26:1324-37.
42. Bishop JM. Molecular themes in oncogenesis. Cell 1991;44:235-48.
43. Rodenhuis S. ras And human tumors. Semin Cancer Biol 1992;3:241-7.
44. Beaupre DM, Kurzrock R. RAS and leukemia: from basic mechanisms to gene-directed therapy. J Clin Oncol 1999;17:1071-9.
45. Davies H, Bignell GR, Cox C, et al. Mutations of the BRAF gene in human cancer. Nature 2002;417:949-54.
46. Frattini M, Ferrario C, Bressan P, et al. Alternative mutations of BRAF, RET and NTRK1 are associated with similar but distinct gene expression patterns in papillary thyroid cancer. Oncogene 2004;23:7436-40.
47. Alt FW, Kellems RE, Bertino JR, Schimke RT. Selective multiplication of dihydrofolate reductase genes in methotrexate-resistant variants of cultured murine cells. J Biol Chem 1978;253:1357-70.
48. Cowell JK. Double minutes and homogeneously staining regions: gene amplification in mammalian cells. Annu Rev Genet 1982;16:21-59.
49. King CR, Kraus MH, Aaronson SA. Amplification of a novel v-erbB-related gene in a human mammary carcinoma. Science 1985;229:974-6.
50. Schwab M, Alitalo K, Klempnauer KH, et al. Amplified DNA with limited homology to myc cellular oncogene is shared by human neuroblastoma cell lines and a neuroblastoma tumour. Nature 1983;305:245-8.
51. Press MF, Bernstein L, Thomas PA, et al. HER-2/neu gene amplification characterized by fluorescence in situ hybridization: poor prognosis in node-negative breast carcinomas. J Clin Oncol 1997;15:2894-904.
52. Ohta M, Inoue H, Cotticelli MG, et al. The human *FHIT* gene, spanning the chromosome 3p14.2 fragile site and renal carcinoma associated translocation breakpoint, is abnormal in digestive tract cancers. Cell 1996;84:587-97.
53. Huebner K, Croce CM. *FRA3B* and other common fragile sites: the weakest links. Nat Rev Cancer 2001;1:214-21.
54. Goldman JM, Melo JV. Targeting the BCR-ABL tyrosine kinase in chronic myeloid leukemia. N Engl J Med 2001;344:1084-6.
55. Heinrich MC, Blanke CD, Druker BJ, Corless CL. Inhibition of KIT tyrosine kinase activity: a novel molecular approach to the treatment of KIT-positive malignancies. J Clin Oncol 2002;20:1692-703.
56. Casali PG, Messina A, Stacchiotti S, et al. Imatinib mesylate in chordoma. Cancer 2004;101:2086-97.
57. Wang ZG, Delva L, Gaboli M, et al. Role of PML in cell growth and the retinoic acid pathway. Science 1998;279:1547-51.
58. Salomoni P, Pandolfi PP. The role of PML in tumor suppression. Cell 2002;108:165-70.
59. Calin GA, Dumitru CD, Shimizu M, et al. Frequent deletions and down regulation of micro-RNA genes miR15 and miR16 at 13q14 in chronic lymphocytic leukemia. Proc Natl Acad Sci U S A 2002;99:15524-9.
60. Calin GA, Sevignani C, Dumitru CD, et al. Human microRNA genes are frequently located at fragile sites and genomic regions involved in cancers. Proc Natl Acad Sci U S A 2004;101:2999-3004.
61. Calin GA, Croce CM. MicroRNA signatures in human cancers. Nat Rev Cancer 2006;6:857-66.
62. Calin GA, Ferracin M, Cimmino A, et al. A microRNA signature associated with prognosis and progression in chronic lymphocytic leukemia. N Engl J Med 2005;353:1793-801. [Erratum, N Engl J Med 2006;355:533.]
63. Yanaihara N, Caplen N, Bowman E, et al. Unique microRNA molecular profiles in lung cancer diagnosis and prognosis. Cancer Cell 2006;9:189-98.
64. Volinia S, Calin GA, Liu C-G, et al. A microRNA expression signature of human solid tumors defines cancer gene targets. Proc Natl Acad Sci U S A 2006;103:2257-61.
65. Nakamura T, Canaani E, Croce CM. Oncogenic All1 fusion proteins target Drosha-mediated microRNA processing. Proc Natl Acad Sci U S A 2007;104:10980-5.
66. Eis PS, Tam W, Sun L, et al. Accumulation of miR-155 and BIC RNA in human B cell lymphomas. Proc Natl Acad Sci U S A 2005;102:3627-32.
67. Costinean S, Zanesi N, Pekarsky Y, et al. Pre-B cell proliferation and lymphoblastic leukemia/high grade lymphoma in E(mu)-miR155 transgenic mice. Proc Natl Acad Sci U S A 2006;103:7024-9.
68. Johnson SM, Grosshans H, Shingara J, et al. RAS is regulated by the let-7 microRNA family. Cell 2005;120:635-47.
69. Cimmino A, Calin GA, Fabbri M, et al. miR-15 And miR-16 induce apoptosis by targeting BCL2. Proc Natl Acad Sci U S A 2005;102:13944-9. [Erratum, Proc Natl Acad Sci U S A 2006;103:2464.]
70. Meng F, Henson R, Lang M, et al. Involvement of human microRNA in growth and response to chemotherapy in human cholangiocarcinoma cell lines. Gastroenterology 2006;130:2113-29.