This is a review article Stan wrote in 1988 while he was researching at Washington University, St. Louis. In it he describes several of the recognized chromosomal and DNA rearrangements within the immunoglobulin synthesis regions that cause and define many white blood cell cancers. The capability to identify the precise molecular genetic changes was available after 1975. The chromosomal and DNA changes described here are used at Barnes (Washington University) Hospital and elsewhere to diagnose, treat, and follow patients with lymphoma and leukemia.

1988

B-Lymphoid Neoplasms: Immunoglobulin Genes as Molecular Determinants of Clonality, Lineage, Differentiation, and Translocation

Stanley J. Korsmeyer, M.D.

Division of Hematology-Oncology, Departments of Medicine and Microbiology-Immunology, Howard Hughes Medical Institute, Washington University School of Medicine, St. Louis, Missouri

Editor's Introduction

The powerful technology of molecular genetics has now revealed the B or T cell origin of lymphoid neoplasms, enabling their classification as distinct maturational stages of B or T cell development. By the ordered sequence of Ig gene rearrangement it has now become clear that acute lymphoblastic leukemia and the lymphoid blast crisis of chronic myelogenous leukemia are developmental stages of B cell precursors, and that the Ig gene rearrangement pattern is unique to each tumor and serves as a marker to follow its course and to search for low levels of residual tumor after therapy. Dr. Stanley Korsmeyer, in his lucid and illuminating review explains the DNA rearrangement of the immunoglobulin and T cell receptor genes that create specific markers capable of establishing the cellular lineage and stage of development of a lymphoid malignancy.

WILLIAM J. HARRINGTON, M.D.

The realization that lymphomas and some leukemias reflected distinct stages of normal lymphocyte development profoundly affected our concepts of their origin and transformation. The generation of monoclonal antibodies to lymphoid cell surface antigens made it possible to classify many of these neoplasms as distinct maturational stages of B- or T-cell development.[1] Despite these advances, it was frequently impossible to determine the B- or T-cell origin or the clonality of a lymphoid neoplasm. A

Adv Intern Med 33:1–16, 1988
© 1988, Year Book Medical Publ[illegible]

number of neoplasms represented cells early in differentiation, prior to the expression of lineage-restricted markers. Other problems included lymphomas possessing large numbers of normal cells admixed with the neoplastic cells. Indeed, the ability to decide that a lymphoid neoplasm was of monoclonal origin was usually limited to mature B-cell malignancies displaying the exclusive presence of cell surface κ or λ immunoglobulin (Ig) light chain.

We will explore the contribution of molecular biology in resolving uncertainties concerning cellular lineage, clonality, stage of development, and pathogenesis of lymphoid neoplasms. This review will focus upon the DNA rearrangements that assemble an Ig gene as a molecular marker for B-cell tumors. Much of our knowledge concerning the basic mechanisms of Ig gene recombination, expression, and selection was gleaned from examining clonal expansions of cells provided by lymphoid tumors. This information indicated an ordered sequence to Ig gene rearrangement and revealed that acute lymphoblastic leukemia (ALL) and the lymphoid blast crisis phase of chronic myelogenous leukemia (CML) were developmental stages of B-cell precursors. Moreover, the Ig gene rearrangement pattern was unique to each tumor and served as a marker to follow its natural history and search for low levels of residual tumor. Some neoplasms of uncertain origin could now be classified by the determination of their gene rearrangement pattern. Moreover, errors were defined in Ig rearrangement and expression that accounted for the truncated peptides of heavy chain disease. Finally, unanticipated rearrangements of the Ig genes have been discovered, which account for disease-associated interchromosomal translocations. This type of rearrangement juxtaposes the Ig locus with a cellular oncogene and directly contributes to the malignant phenotype.

Immunoglobulin Gene Subsegments Recombine to Generate Antibody Diversity

Humans can generate a seemingly unlimited number of different antibody specificities (10^6–10^9).[2,3] The antibody molecule is composed of two identical heavy chains that can theoretically assort with any two identical light chain molecules. The antigen recognition portion of the molecule is located in the N terminal variable (V) portion of the molecule, while the C terminal constant (C) portion is invariant and performs effector functions. This unusual immunoglobulin molecule is encoded by multiple gene subsegments, as Dreyer and Bennett first proposed.[4] Within their germline or embryonic form, Ig genes are organized as discontinuous pieces of separated DNA that are assembled by a process of DNA rearrangement (Fig 1). The variable portion of the human heavy chain gene is generated from families of variable (V_H) regions, a set of internal diversity (D_H) regions, and a strip of

six alternative joining (J_H) gene segments located on chromosome segment 14q32.[7,8] During the first step of Ig gene assemblage, a D_H region recombines with a J_H region, and then a V_H region fuses with that D_H/J_H combination (see Fig 1). Most human B cells (85%) have needed to rearrange both Ig alleles in order to create a functional $V_H/D_H/J_H$ recombination and thus possess one invalidly rearranged H-chain allele. Initially, B cells utilize the most proximal located C_H region, Cμ, and produce only IgM. Later they may utilize an alternative splicing mechanism to place either Cμ or Cδ with the same V/D/J and display simultaneous surface IgM and IgD.[9,10] Finally, a process of H-chain class switch may occur to move a more distal located constant region of Cγ, Cε, or Cα next to the same V/D/J (see Fig 1).[11] This class switch is a second form of DNA rearrangement mediated by repetitive elements known as switch sites, which precede these C_H regions.[12,13] The H-chain class switch results in the production of IgG, IgA, or IgE in which the same antigen specificity of the assembled variable region is now linked to a constant region that provides a different physiologic function to the molecule.

Mature B cells produce but one of the two available light chain classes, κ or λ. Moreover, each cell manufactures only the maternal or paternal allele of that light chain, a process known as allelic exclusion. The human κ chain gene is located on chromosome segment 2p11 and consists of separate Vκ and Jκ segments that are recombined as Vκ/Jκ junctures and expressed with a single Cκ gene.[14,15] In contrast, the λ locus on chromosome segment 22q11 is comprised of multiple Jλ—Cλ clusters (6 to 9 in number) and upstream Vλ segments.[16] Similarly however, a Vλ/Jλ rearrangement occurs to activate this locus.

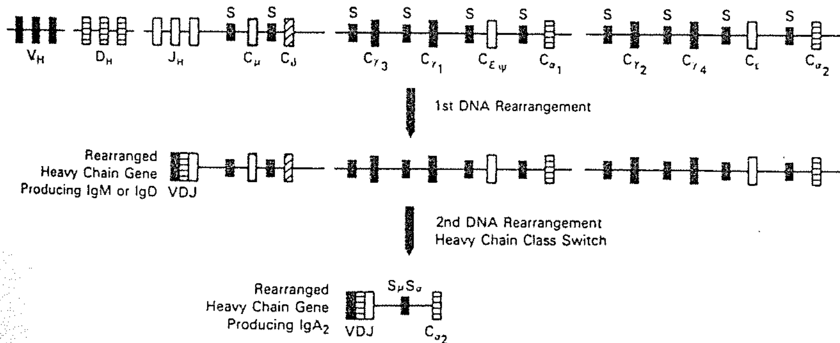

FIG 1.
Embryonic/germline human heavy chain gene (14q 32) locus. The first DNA rearrangement assembles a variable (*V_H*), diversity (*D_H*), and joining (*J_H*) region to complete the variable portion of the molecule. Subsequently, a second DNA rearrangement between homologous switch sites *(S)* can occur, resulting in a heavy chain class switch that moves a more distal constant region next to the assembled V/D/J.

Rearrangement of Immunoglobulin Genes as Clonal Markers

The molecular event that moves and combines a V and J segment results in the relocation of restriction endonuclease sites at the DNA level. Thus, the DNA restriction fragment that bears an Ig gene in its rearranged form is of altered size compared with the germline form of the gene (Fig 2). Because B-cell malignancies represent clonal expansions of a single progenitor cell, each tumor will possess multiple copies of an identical V/J type rearrangement. This expanded copy number makes it possible to detect such rearrangements by routine Southern blots and has facilitated the molecular cloning of these recombinations (see Fig 2). Because each individual B cell will use only one V, one D, and one J of many possibilities, the pattern of gene rearrangement will be unique to that cell. Thus, these rearrangements serve as tumor-associated molecular markers useful in determining clonality and lineage commitment.[17]

Non-T, Non-B Cell Acute Lymphoblastic Leukemia: A Developmental Series of B-Cell Precursors

Most (80%) ALLs were of uncertain cellular origin prior to the era of gene analysis, because they lacked T-cell–associated surface antigens and also failed to display cell surface Ig. Therefore, they had been referred to as non-T, non-B, or null cell ALL. Analysis of their Ig genes revealed that ALLs displayed a developmental cascade of gene rearrangements.

Almost all of these cells demonstrated immunoglobulin heavy chain gene rearrangement, and 60% retained germline κ and λ light chain genes (Fig 3).[18,19] The 40% that had moved on to the later event of light chain gene rearrangement did so in a κ before λ order. Cells that possessed κ gene rearrangements, retained germline λ genes. In contrast, those with λ gene rearrangement had no remaining germline κ genes, having usually deleted them. In fact, these Alls provided clonal expansions of pre-B cells that permitted the cloning of the κ deleting element (κde).[20] The κde is responsible for the loss of κ genes that precedes λ gene rearrangement. Overall, the Ig gene pattern in the pre-B form of ALL indicated a hierarchy in which heavy chain rearrangement preceded light, and κ rearranged before λ (see Fig 3).

A coordinate sequence of cell-surface antigen expression accompanies the cascade of gene rearrangements (see Fig 3).[19,21] The earliest B-cell precursors display heavy chain rearrangements and cell-surface histocompatibility antigens (HLA)-DR, and B_4 (a B-cell associated antigen). Next, the common acute lymphoblastic leukemia antigen (CALLA) is added, and light chains subsequently rearrange. The timing of B1 antigen addition varies. The J-chain glycopeptide that ultimately connects pentameric IgM or dimeric IgA may also be expressed as early as a pre-B cell stage.[22]

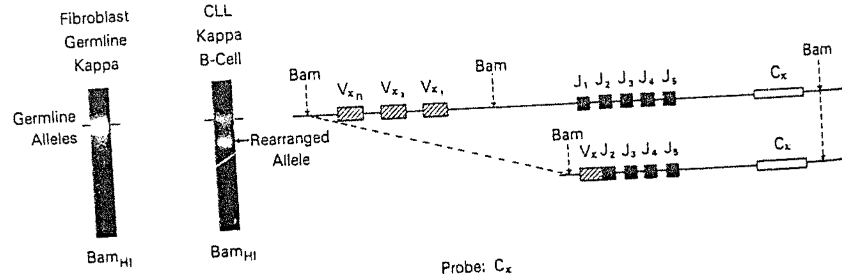

FIG 2.
Detection of rearranged human kappa gene allele. The upper schematic represents a germline K gene in which a C_K probe recognizes a 12.0-Kb BamHI fragment. With activation of this allele, one of many V_K segments is juxtaposed with one of five alternative J_K segments. Rearrangement introduces a new 5′ BamHI restriction fragment. The Southern blot reveals that a rearranged K allele occupies an altered

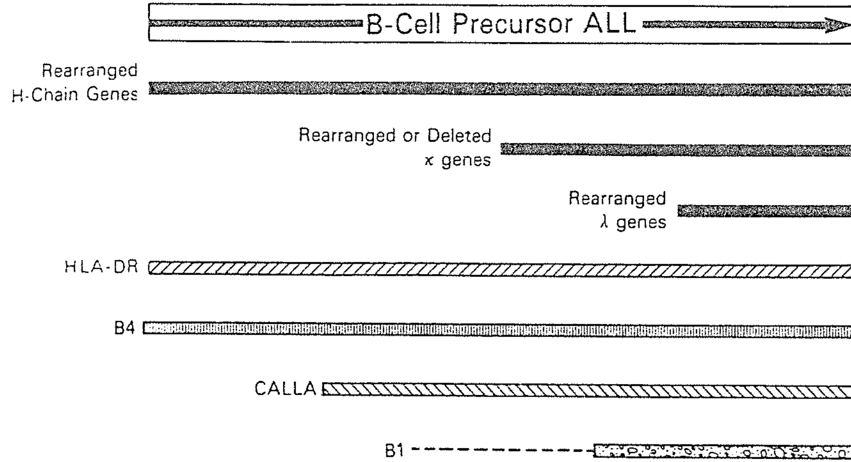

FIG 3.
The non-T, non-B acute lymphoblastic leukemias represent a developmental cascade of B-cell precursors. Heavy *(H)* chain rearrangements precede light chain, and κ precede λ. A coordinate sequence of surface antigen expression occurs in which *HLA-DR* and *B4* precede the common acute lymphoblastic leukemia antigen *(CALLA)* and *B1,* which has a variable time of onset.

Some Pre-B ALLs Display Clonal Heterogeneity Over Time

B-cell precursor ALLs by definition lack surface immunoglobulin. In addition, the surface antigens they express are found on normal pre-B cells.

For this reason we turned to their Ig gene rearrangement pattern as a tumor-specific marker to follow the natural history of such ALLs.[23] We noted that most patients (11 of 15) had identical gene patterns at diagnosis and subsequent relapses indicating that all leukemic episodes represented the same clone. The four patients that displayed clonal variations in Ig genes over time always shared at least one identical Ig gene rearrangement. Thus, all episodes of leukemia in a single patient arose from a common clonal progenitor cell. None of these leukemias were really biclonal in that they did not arise from separate transformation events. However, just as pre-B ALL as a collective group demonstrates a developmental series of cells, individual cases can evolve over time.

Identifying Minimal Residual Disease by Immunoglobulin Gene Rearrangements

The stochastic nature of Ig gene rearrangement creates a molecular marker unique to that cell. In order for a DNA rearrangement to be detected by Southern blot analysis, however, it must be present in 1% to 5% of all cells present. Thus, Ig gene rearrangement within a tissue represents a very specific and a moderately sensitive measure of clonal cells. We utilized gene rearrangement to look for residual leukemia 28 days following the induction of chemotherapy.[23] There was a perfect correlation with the histopathologic identification of lymphoblasts and the molecular detection of clonality. Those patients without lymphoblasts displayed no rearrangements, indicating that leukemic cells were actually eliminated rather than morphologically altered. Several bone marrow aspirates obtained during remission revealed molecular clonality in the absence of lymphoblasts, and these patients subsequently relapsed. Moreover, significant negatives were noted. Several patients with equivocal evidence for disease (1% to 6% lymphoblasts, sheets of lymphocytes, scattered islands of lymphoblasts) had entirely germline gene configurations, and none of these patients has subsequently relapsed.[23] The examination of T-cell receptor genes, chromosomal translocation breakpoints, in addition to Ig genes will place DNA markers on a large number of neoplasms and expand this approach for looking for minimal residual disease.

Pre-B Cell Lymphoid Blast Crisis in CML

The clonal progenitor cell of CML is apparently a rather pluripotential stem cell. Assessment of G-6-PD alleles and the t(9;22) Philadelphia chromosomal marker has indicated that numerous hematopoietic lineages can be clonally affected.[24] Because of the multipotential involvement of this dis-

ease, the precise lineage identity of the blast crisis phases were often uncertain. The finding that Ig heavy and at times light chains were rearranged in CML lymphoid blast crisis indicated that they were genetically committed B-cell, precursors.[25, 26] In fact, the clonally committed cells in CML proved capable of differentiation. For example, single patients may demonstrate clinically separate lymphoid blast crisis episodes over time. When such a case was molecularly examined, both lymphoid crisis arose from a common lymphoid progenitor cell that possessed identical heavy chain rearrangements. However, differentiation appeared to be intact in these leukemic cells, because one of the lymphoid episodes had progressed to light chain gene rearrangement.[26] Overall, most of the lymphoid blast crisis events are pre-B cell in type, although a rare T-cell blast crisis has been described.[27] The t(9;22)(q34, q11) Philadelphia translocation results in a fusion gene and fusion protein between the c-Abl oncogene from chromosome 9 and the BCR gene on chromosome 22.[28] The resultant fusion peptide (p210) has an enhanced tyrosine kinase activity and might conceivably favor the B-cell developmental pathway.[29]

Immunoglobulin Gene Configuration in Mature B-Cell Malignancies

Mature B cells must possess the obligate rearrangement of heavy and light chain immunoglobulin genes corresponding to their cell-surface antibody. In addition to the initial DNA rearrangement to assemble the V/D/J of the heavy chain variable region, such cells may display a second rearrangement known as the heavy chain class switch (see Fig 1). This allows the production of the more 3′ constant region responsible for IgG, IgA, or IgE. Mature B cells also possess archeologic evidence of the ordered rearrangement of light chain genes. In general, κ-producing cells retain germline λ genes, while λ-producing cells have usually deleted their κ genes.[30] These molecular events represent further clonal markers that testify to the extent of genetic maturation.

This background of information has been of utility when defining the cellular origin of neoplasms of uncertain cell type. One such leukemia, hairy-cell leukemia, had been ascribed the properties of B, T, or macrophages by various investigators. However, an examination of Ig genes revealed rearrangement and expression patterns characteristic of mature B cells.[31] Of interest, nearly all B-cell type hairy-cell leukemias display a cell-surface receptor (Tac) for interleukin-2 (T-cell growth factor).[31] This finding in human lymphoid neoplasms promoted research revealing that activated normal B cells also express interleukin-2 receptors.[32] Thus, hairy-cell leukemia may represent the transformation of a normal counterpart: activated B cells.

Gene Defects in Human Heavy Chain Disease

A subset of B-cell lymphoproliferative disorders display only surface Ig heavy chain and no associated light chain.[33] Furthermore, the heavy chains are abnormally truncated in size. Protein studies indicated that these heavy chains lacked all or most of the variable region and often began at the sites of constant region domains. One could envision three general forms of molecular defects that could account for these shortened heavy chains: (1) postsynthetic degradation; (2) DNA deletion; (3) RNA-splicing abnormality. The initially synthesized proteins in heavy chain disease (HCD) are always small, indicating that no extensive protein degradation occurs and implicating DNA or RNA level defects.

A number of different defects in the complex process of correctly expressing a heavy chain are proving to account for HCD. Mistakes in V/D/J rearrangement or in heavy chain class switch might delete important areas and result in an incorrectly initiated RNA or an RNA-splicing abnormality. Alternatively, somatic mutation could also alter the donor or acceptor splice sites and thus eliminate the corresponding variable and constant region subsegments. We observed that a μ HCD protein arose from an aberrant RNA splice between the leader sequence and the Cμ region (Fig 4).[34] An unexpected DNA insertion/deletion event eliminated a J_H donor splice site and forced the leader to be juxtaposed with the Cμ region. This molecular defect accounted for the truncated μ chain that lacked variable information and lacked associated light chains. Gamma-type heavy chain diseases have also been analyzed and possess abnormal DNA rearrangements that remove portions of their variable regions.[35, 36] In addition, an abnormal heavy chain class switch has been implicated in removing the portions of the Cγ region that are missing in the protein. Overall, a variety of defects at a number of the multisteps of Ig gene assembly and expression can account for the proteins of HCD.

T-Cell Receptor (TCR) Genes

T cells also demonstrate antigen-specific recognition through a cell surface T_3-Ti (idiotype) complex. The Ti portion is the antigen receptor and is comprised of α and β TCR peptides. The genes for both the α and β chain of the TCR have been cloned and characterized.[37–39] Like the Ig gene, the TCR genes also rearrange to assemble. The β TCR is located at chromosome segment 7q35 and bears only an evolutionary homology to the Ig locus. It has two available constant regions (Cβ1) and (Cβ2), sets of diversity (Dβ) and joining (Jβ) segments as well. The α TCR gene is located at 14q11 and has a single constant region (Cα), a very long stretch of joining (Jα) segments, and many variable (Vα) regions. These genes have rearranged in mature T cells to produce the α, β chain heterodimer that is complexed with cell-surface T_3. In addition, there is a third TCR

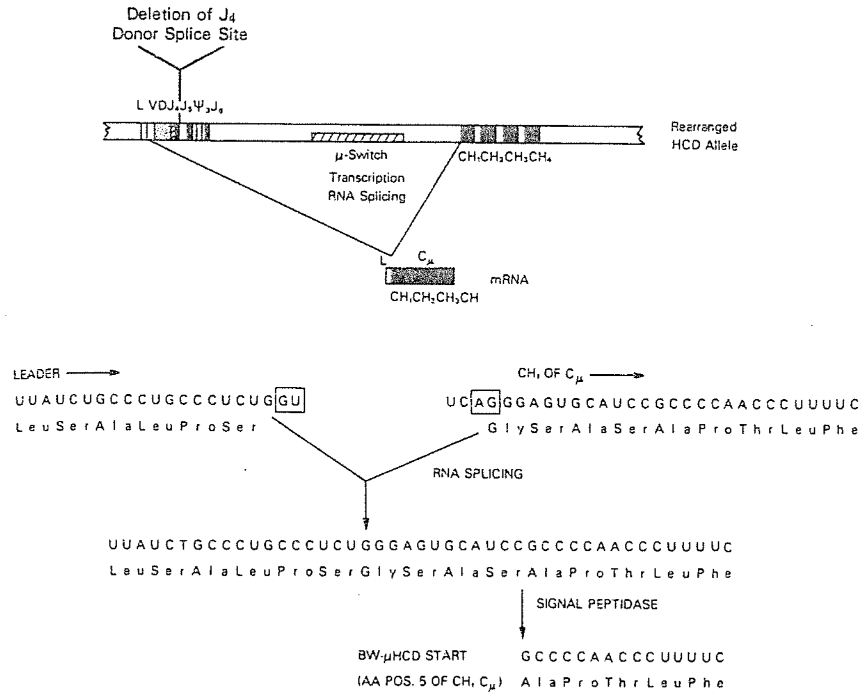

FIG 4.
A small DNA deletion eliminates the J_4 donor splice site of a patient with a μ heavy chain disease. This results in an aberrant splice between the leader *(L)* and Cμ region. A new recognition amino acid sequence for signal peptidase is created and results in a truncated μ chain that begins at amino acid 5 of CH_1 *of* Cμ.

pressed early in T-cell development and later is present on only a minority of cells bearing a T_3 molecule.

T-cell receptor gene rearrangement provided the sorely needed marker that proved most T-cell malignancies to be of clonal origin. Furthermore, analysis of TCR genes has demonstrated clonality in some T-cell disorders such as the T_8 lymphocytosis with associated hematocytopenias that were not previously known to malignant.[41] While the demonstration of clonality is not tantamount to a verdict of malignancy nor necessarily a warrant for therapy, it does indicate the single-cell origin of these disorders.

Immunoglobulin and T-Cell Receptor Rearrangements Spillover to the Opposite Lineage

While rearrangement of α and β TCR genes are required in mature T cells, we have noted that α, β and γ TCR activation may occur at times in B

cells.[41, 42] Moreover, approximately 10% of bona fide T cells and occasionally myeloid cells will display an Ig heavy chain rearrangement.[17, 19, 42] Fortunately, the later developmental event of Ig light chain rearrangement is restricted to the B series. This lineage crossover of antigen receptor rearrangements is seen within normal as well as malignant cells. This may reflect the fact that a single recombinase enzyme mediates rearrangement of both the Ig and TCR loci. Such rearrangements may occur early in development before full lineage commitment has occurred. Consistent with this notion, early B-cell precursor forms of ALL have a higher incidence of β and γ TCR rearrangement (20% and 45%) than more mature B cells.[42] Because of the lineage spillover of TCR and Ig gene rearrangement, caution must be used when applying such markers to determine cellular type. The presence of Ig κ or λ light chain rearrangement is strong evidence of a B-cell commitment. Similarly, the presence of TCR activation in the absence of Ig gene rearrangement is conclusive for a T-cell origin. With these general guidelines in place, the application of Ig and TCR rearrangements may frequently resolve the cell type of lymphoid biopsy tissues containing large numbers of admixed normal cells. In this setting the truly neoplastic cells of origin will display clonal rearrangements, while the infiltrating normal cells will be polyclonal in nature.

Chromosomal Translocations Occur at the Immunoglobulin Gene Loci

Distinct interchromosomal translocations are uniquely or frequently found with specific types of neoplasms. B-cell tumors possess chromosomal translocations at the exact cytogenetically defined bands that possess the Ig genes.[43, 44] Indeed, in Burkitt's lymphoma the exact site of chromosomal juncture landed within the Ig gene loci. The most frequent translocation in Burkitt's lymphoma t(8;14) usually breaks at the μ switch region or the joining (J_H) region of the heavy chain gene at 14q32.[45] The variant 10% of Burkitt's that break elsewhere choose to use the site of the Ig light chain gene, κ at 2p11 or λ at 22q11.[46] The other partner in this reciprocal translocation was chromosome 8 at band q24, the site of the c-myc cellular oncogene (Fig 5). The introduction of c-myc into the foreign location of an immunoglobulin locus leads to its deregulation during the cell cycle and presumably results in a transformation event.

Chromosome segment 14q32 is repeatedly associated with sites of translocation in mature B-cell neoplasms. A translocation between the heavy chain locus and a putative gene at 11q13, Bcl-1 is found frequently in multiple myeloma and occasionally in other B-cell tumors.[47] Similarly, human follicular lymphomas possess a t(14;18) (q32;q21) translocation that is present in perhaps 85% of small-cell type follicular lymphomas. We and others have cloned this chromosomal breakpoint and characterized a

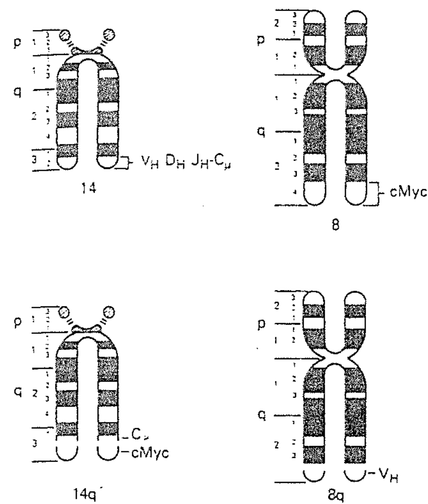

FIG 5.
Burkitt's lymphoma t(8;14). A normal chromosome 14 possesses the effective $V_H/D_H/J_H$ gene rearrangement responsible for IgM production. The normal chromosome 8 retains a nonexpressed copy of *cMyc*. The *14q+* chromosome has received a portion of chromosome 8 bearing the *cMyc* genes. The *8q−* chromosome receives a portion of chromosome 14, including some V_H regions.

new B-cell proliferation-associated gene at 18q21, Bcl-2.[48, 49] The chromosome 14 side of the breakpoint involves the 5′ end of a J_H region on the derivative (der) 14 chromosome ($14q^+$) and the 3′ end of a D_H region on the der (18) ($18q^-$) (Fig 6). Extra nucleotides that are neither of chromosome 14 nor 18 origin are found at both chromosomal breakpoints and are reminiscent of the "N" segment additions normally inserted at sites of V/D and D/J juncture during normal Ig assembly. These findings indicate that the chromosomal breakage on 14 is mediated by the immunoglobulin recombinase enzyme. Furthermore, this indicates that the t(14;18) actually occurs at a pre-B cell stage of development during the first step of Ig gene joining, the attempted D_H to J_H recombination. Despite this, the final tumor is a mature B cell. Thus, a transformation event occurs much earlier in development than the final phenotypic stage of the ultimate tumor. The breaks on chromosome segment 18q21 are focused in a 2.8-Kb major breakpoint region (70%), and most of these fall in a small 150-bp cluster region. The mechanism of breakage on 18q21 appears to be a naturally occurring staggered dsDNA break unrelated to Ig recombinase. The break on 18 interrupts the 3′ exon of the Bc1-2 gene and places it in the same transcriptional orientation as the Ig gene locus. This results in a Bc1-2-Ig fusion RNA transcript. However, the portion of the messenger RNA encoding the BC1-2 protein product is not interrupted. Instead, this event results in a deregulation and overexpression of this gene in the follicular lymphomas.

The clustering of chromosomal breakpoints allows the placement of a unique rearrangement marker on each chromosomal translocation. This allows the translocation event to be followed throughout the course of the tumor. Such studies revealed that the translocation breakpoint remains un-

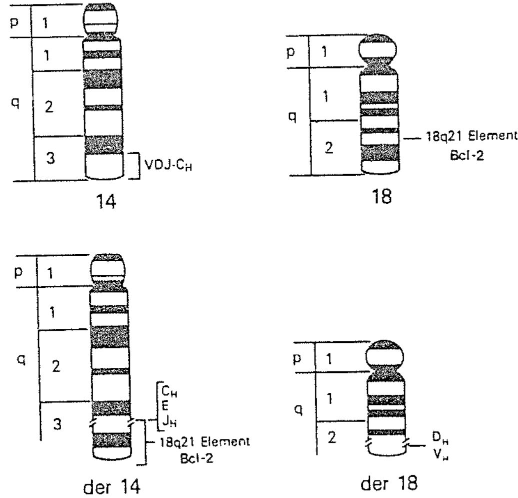

FIG 6.
Follicular lymphoma with t(14;18)(q32;q21) translocation. The normal chromosome 14 contains a V/D/J responsible for immunoglobulin production. The derivative 14 *(der 14)* chromosome has introduced a new B-cell associated transforming gene into J_H near the enhancer *(E)* element. The reciprocal *der 18* partner has received D_H and V_H portions of chromosome 14.

changed throughout the disease, while the normal Ig genes may undergo secondary rearrangements.[51] These result from evolving tumor heterogeneity, and not the biclonal emergence of new clones. Moreover, the cloning of such breakpoints allows the molecular refinement of cytogenetic classification. We have found t(14;18) rearrangements at the gene level that were missed, internally hidden, or indeterminant by routine cytogenetics.[52] The molecular definition of chromosomal translocations has defined new potential transforming genes and also provided important tools in improving our understanding of tumor behavior.

Summary

The basic rules of Ig gene recombination that were learned from examining clonal proliferations of B cells have paid enormous dividends in improving our understanding of B-cell malignancy. The DNA rearrangements of Ig genes creates a tumor-specific marker capable of establishing the clonality, cellular lineage, and stage of development of a lymphoid tumor. Most importantly, the Ig genes have proved to be the sites of interchromosomal translocations that contribute directly to the malignant phenotype. These genetic tools hold the promise of improving our classification schemes, providing sensitive and specific approaches to following the clinical course, and providing insights into pathogenesis that will prompt improved therapies.

References

1. Aisenberg AC: Cell surface markers in lymphoproliferative disease. *N Engl J Med* 1978; 304:331.
2. Leder P: The genetics of antibody diversity. *Sci Am* 1982; 246:102.
3. Tonegawa S: Somatic generation of antibody diversity. *Nature* 1983; 302:575.
4. Dreyer WJ, Bennett JC: The molecular basis of antibody formation: A paradox. *Proc Natl Acad Sci USA* 1965; 54:864.
5. Brack C, Hirama M, Lenhard-Schuller R, et al: A complete immunoglobulin gene is created by somatic recombination. *Cell* 1978; 15:1.
6. Seidman JG, Leder P: The arrangement and rearrangement of antibody genes. *Nature* 1978; 276:790.
7. Siebenlist U, Ravetch JV, Korsmeyer SJ, et al: Human immunoglobulin D segments encoded in tandem multigenic families. *Nature* 1981; 294:631.
8. Ravetch JV, Siebenlist U, Korsmeyer SJ, et al: The structure of the human immunoglobulin μ locus: Characterization of embryonic and rearranged J and D genes. *Cell* 1981; 27:583.
9. Knap MR, Liu CP, Newell N, et al: Simultaneous expression of immunoglobulin μ and heavy chains by a cloned B cell lymphoma: A single copy of the V_H gene is shared by two adjacent C_H genes. *Proc Natl Acad Sci USA* 1982; 79:2996.
10. Moore KW, Rogers J, Hunkapiller T, et al: Expression of IgD may use both DNA rearrangement and RNA splicing mechanisms. *Proc Natl Acad Sci USA* 1981; 78:1800.
11. Davis MM, Kim SK, Hood LE: DNA sequences mediating class switching α-immunoglobulins. *Science* 1980; 209:1360.
12. Kataoka T, Miyata T, Honjo T: Repetitive sequences in class-switch recombination regions of immunoglobulin heavy chain genes. *Cell* 1981; 23:357.
13. Ravetch JV, Kirsch IR, Leder P: Evolutionary approach to the question of immunoglobulin heavy chain switching: Evidence from cloned human and mouse genes. *Proc Natl Acad Sci USA* 1980; 77:6734.
14. Hieter PA, Max EE, Seidman JG, et al: Cloned human and mouse kappa immunoglobulin constant and J region genes conserve homology in functional segments. *Cell* 1980; 22:197.
15. Malcolm S, Barton P, Murphy C, et al: Localization of human immunoglobulin κ light chain variable region genes to the short arm of chromosome 2 by in situ hybridization. *Proc Natl Acad Sci USA* 1982; 79:4957.
16. Hieter PA, Hollis GF, Korsmeyer SJ, et al: Clustered arrangement of immunoglobulin λ constant regions genes in man. *Nature* 1981; 294:536.
17. Arnold A, Cossman J, Bakhshi A, et al: Immunoglobulin gene rearrangements as unique clonal markers in human lymphoid neoplasms. *N Engl J Med* 1983; 309:1593.
18. Korsmeyer SJ, Hieter PA, Ravetch JV, et al: Developmental hierarchy of immunoglobulin gene rearrangements in human leukemic pre-B cells. *Proc Natl Acad Sci USA* 1981; 78:7096.

19. Korsmeyer SJ, Arnold A, Bakhshi A, et al: Immunoglobulin gene rearrangements and cell surface antigen expression in acute lymphocytic leukemias of T cell and B cell precursor origins. *J Clin Invest* 1983; 71:301.
20. Siminovitch KA, Bakhshi A, Goldman P, et al: A uniform deleting element mediates the loss of κ genes in human B cells. *Nature* 1985; 316:260.
21. Nadler LM, Korsmeyer SJ, Anderson KC, et al: The B cell origin of non-T acute lymphoblastic leukemia: A model for discrete stages of neoplastic and normal pre-B cell differentiation. *J Clin Invest* 1984; 74:332.
22. Max EE, Korsmeyer SJ: The human J chain Gene: Structure and expression in B lymphoid cells. *J Exp Med* 1985; 161:832.
23. Wright JJ, Poplack DG, Bakhshi A, et al. Gene rearrangements as markers of clonal variation and minimal residual disease in acute lymphoblastic leukemia. *J Clin Oncol* 1987; 5:735.
24. Barr RD, Fialkow PJ: Clonal origin of chronic myelocytic leukemia. *N Engl J Med* 1973; 289:307.
25. Ford AM, Molgaard HV, Greaves MF, et al: Immunoglobulin gene organization and expression in hematopoietic stem cell leukemia. *EMBO J* 1983; 2:997.
26. Bakhshi A, Minowada J, Arnold A, et al: Lymphoid blast crises of chronic myelogenous leukemia represent stages in the development of B-cell precursors. *N Engl J Med* 1983; 309:826.
27. Chan LC, Furley AJ, Ford AM, et al: Clonal rearrangement and expression of T cell receptor β gene and involvement of the breakpoint cluster region in blast crisis of CGL. *Blood* 1986; 67:533.
28. Shtivelman E, Lifshitz B, Gale RP, et al: Alternative splicing of RNAs transcribed from the human Abl Gene and from the bcr-abl fused gene. *Cell* 1986; 47:277.
29. Konopka JB, Watangbe SM, Witte ON: An alternation of the human c-abl protein in K562 leukemia cells unmasks associated tyrosine Kinase activity. *Cell* 1984; 37:1035.
30. Hieter PA, Korsmeyer SJ, Waldman TA, et al: Human immunoglobulin κ light chain genes are deleted or rearranged in λ-producing B cells. *Nature* 1981; 290:368.
31. Korsmeyer SJ, Greene WC, Cossman J, et al: Rearrangement and expression of immunoglobulin genes and expression of Tac antigen in hairy cell leukemia. *Proc Natl Acad Sci USA* 1983; 80:4522.
32. Waldmann TA, Goldman CK, Robb RJ, et al: Expression of interleukin-2 receptors on activated human B-cells. *J Exp Med* 1984; 160:1450.
33. Seligman M, Mihaesco E, Preud'homme JL, et al: Heavy chain disease: Current findings and concepts. *Immunol Rev* 1979; 48:145.
34. Bakhshi A, Guglielmi P, Siebenlist U: A DNA insertion/deletion necessitates an aberrant splice accounting for a human mu heavy chain disease protein. *Proc Natl Acad Sci USA* 1986; 83:2689.
35. Alexander A, Steinmetz A, Barritault D, et al: γ heavy chain disease in man: cDNA sequence supports partial gene deletion model. *Proc Natl Acad Sci USA* 1982; 79:3260.
36. Guglielmi P, Bakhshi A, Miahesco E, et al: DNA deletion in human γ heavy chain disease. *Clin Res* 1984; 32:348A.
37. Yanagi Y, Yoshikai Y, Leggett K, et al: A human T-cell specific cDNA clone encodes a protein having extensive homology to immunoglobulin chains. *Nature* 1984; 308:145.

38. Hedrick SM, Cohen DI, Nielson EA, et al: Isolation of cDNA clones encoding T-cell specific membrane-associated proteins. *Nature* 1984; 308:149.
39. Yoshikai Y, Clark SP, Taylor S, et al: Organization and sequences of the variable, joining and constant region genes of the human T-cell receptor α-chain. *Nature* 1985; 316:837.
40. Hayday AC, Saito H, Gillies SD, et al: Structure, organization and somatic rearrangement of T-cell gamma genes. *Cell* 1985; 40:259.
41. Waldmann TA, Davis MM, Bongiovanni KF, et al: Rearrangements of genes for the antigen receptor on T cells as markers of lineage and clonality in human lymphoid neoplasms. *N Engl J Med* 1985; 313:776.
42. Felix CA, Wright JJ, Poplack DG, et al: T cell receptor α, β, and γ genes in T cell and Pre B cell acute lymphoblastic leukemia. *J Clin Invest* 1987 (in press).
43. Rowley JD, Testa JR: Chromosome abnormalities in malignant hematologic disease. *Adv Cancer Res* 1982; 36:103.
44. Yunis JJ: Chromosomal basis of human neoplasia. *Science* 1983; 221:227.
45. Taub R, Kirsch I, Morton C, et al: Translocation of the c-*myc* gene into the immunoglobulin heavy chain locus in human Burkitt lymphoma and murine plasmacytoma cells. *Proc Natl Acad Sci USA* 1982; 79:7837.
46. Lenoir GM, Preud'homme JL, Bernheim A, et al: Correlations between immunoglobulin light chain expression and variant translocation in Burkitt's lymphoma. *Nature* 1982; 298:474.
47. Tsujimoto Y, Yunis JJ, Onorato-Showe L, et al: Molecular cloning of the chromosomal breakpoints of B cell leukemias and lymphomas with the t(11;14) chromosomal translocation. *Science* 1984; 224:2403.
48. Tsujimoto Y, Finger LR, Yunis JJ, et al: Cloning the chromosome breakpoint of neoplastic B cells with the t(14;18) chromosome translocation. *Science* 1984; 226:1097.
49. Bakhshi A, Jensen JP, Goldman PG: Cloning the chromosomal breakpoint of t(14;18) human lymphomas: Clustering around J_H on chromosome 14 and near a transcriptional unit on 18. *Cell* 41:889.
50. Bakhshi A, Wright JJ, Graninger W, et al: Mechanism of the t(14;18) translocation: Structural analysis of both derivative 14 and 18 reciprocal partners. *Proc Natl Acad Sci USA* 1987; 84:2396.
51. Raffeld M, Wright JJ, Lipford E, et al: Clonal evolution of t(14;18) follicular lymphomas demonstrated by immunoglobulin genes and an 18q21 breakpoint region. *Cancer Res* 1987; 47:2537.
52. Lipford E, Wright JJ, Urba W, et al: Refinement of lymphoma cytogenetics by the 18q21 major breakpoint region. *Clin Res* 1986; 34:565A.