Genomic Structure of the Human Ig\(\lambda 1\) Gene Suggests That It May Be Expressed as an Ig\(\lambda 14.1\)-like Protein or as a Canonical B Cell Ig\(\lambda \) Light Chain: Implications for Ig\(\lambda \) Gene Evolution

By Robert J. Evans and Gregory F. Hollis

Summary

In pre-B cells, immunoglobulin \(\mu\) (Ig\(\mu\)) is associated with pre-B cell–specific proteins to form a multimeric complex that is found on the cell surface. One of these proteins is encoded by the three exon Ig\(\lambda\)-like gene 14.1, whose expression is restricted to pre-B cells and occurs from an unrearranged gene. A comparison of the 14.1 gene structure to the seven-gene human Ig\(\lambda\) locus revealed that the most 5' gene, Ig\(\lambda 1\), is organized in a three-exon structure very similar to the 14.1 gene. Transcription and splicing of these three-exon sequences would lead to an mRNA with an open reading frame which could encode a light (L) chain–like protein with a molecular weight of 23,045. Our analysis suggests that two transcripts may be produced from the Ig\(\lambda 1\) gene that share the same Ig\(\lambda 1\) constant region–containing third exon. One transcript would include all three 14.1-related exons and be expressed from the germline gene, and the second transcript would be produced after variable-joining (VJ) recombination has occurred to Ig\(\lambda 1\) and would encode a classic Ig\(\lambda\) L chain protein. The conservation of the genomic organization of the human 14.1 and Ig\(\lambda 1\) genes and the mouse homolog, A\(5\), relative to the classic Ig\(\lambda\) L chain genes provides insight into the evolution of Ig genes.

The development of B cells from stem cells to mature Ig-secreting B cells can be classified by the rearrangement and expression of Ig \(\kappa\) and L chain genes. In this progression, Ig\(\mu\) H chain undergoes VDJ rearrangement, is expressed first, and in part defines the pre-B cell stage of development. Subsequently, the Ig L chain genes, \(\kappa\) and \(\lambda\), undergo rearrangement and expression. Expression of both Ig\(\mu\) and L chain proteins leads to the formation of the Ig tetramer \(\mu2\lambda2\) (\(\lambda\) or \(\lambda\) chain) and marks the transition of pre-B cells to the B cell stage of development (1–6).

During our studies of the human Ig\(\lambda\) locus, we identified and isolated two clones, 14.1 and 16.1, based on their shared homology with the Ig\(\lambda\) C region (7, 8). More recently, we have shown that at least one of these clones, 14.1, is expressed as a 1-kb transcript exclusively in pre-B cells (9). This transcript contains both Ig\(\lambda\) J and C region homologies as well as 5' sequence that is not derived from an Ig V region.

Sequence analysis of a cDNA clone, Hom-1, derived from the 14.1 gene revealed a long open reading frame capable of encoding a protein of an unprocessed mol wt of 22,944. Polyclonal antisera, generated to a peptide predicted by the nucleotide sequence, identified a 22-kD protein as the product of the 14.1 gene. Immunoprecipitation experiments using anti-Ig\(\mu\) antisera demonstrated that at least two proteins, of 16 and 22 kD, are associated with Ig\(\mu\) in pre-B cells.

Taken together, the immunoprecipitation and Western analysis suggested that the protein product encoded by the 14.1 gene is complexed to Ig\(\mu\) in pre-B cells (9). Recently, these results have been confirmed and extended by Kerr et al. (10) who have shown that cell surface Ig\(\mu\) in pre-B cells is covalently linked to two protein chains, 16 and 22 kD in size, that crossreact with antisera directed against human Ig\(\mu\) protein.

Similar results have been described for mouse pre-B cells where a surrogate L chain, termed \(\omega\), has been shown to be complexed with Ig\(\mu\) to form a \(\mu2\omega2\) tetramer (11). In separate experiments, a cDNA encoding a pre-B cell–specific transcript, A\(5\), was isolated that contains homology to Ig J and C regions (12–14). The mouse A\(5\) gene contains the putative coding sequence for the mouse \(\omega\) protein, and based on sequence similarity, is likely to be the mouse homologue of the human 14.1 gene (9, 15).

Analysis of the 14.1 gene indicates that it is expressed without gene rearrangement and is encoded in three exons, in which exons 2 and 3 contain Ig J and C region homology, respectively. Characterization of the genomic structure of the 14.1 gene led to the discovery that the human Ig\(\lambda 1\) gene, the most 5' gene of the seven gene \(\lambda\) locus, is organized in a similar three-exon structure. This germline Ig\(\lambda 1\) gene contains appropriate RNA processing signals that could lead to
the production of a transcript with one long open reading frame with an in frame AUG start site. This analysis suggests that the IgM gene may be expressed in two ways. One transcript would be expressed from the germline gene and include all three 14.1-related exons and the second transcript would be produced from a rearranged IgM gene after V-J recombination and would encode a classic IgM L chain protein.

Materials and Methods

**DNA and RNA Analysis.** Plasmid DNAs (1 µg/lane) were digested with the indicated restriction enzyme according to manufacturers recommendations, fractionated on a 0.8% agarose gel, and transferred to nitrocellulose paper (16). RNA extraction and blots were performed as described (9, 17). DNA and RNA blots were hybridized to the following 32P-labeled human DNA probes: (a) 847-bp fragment including all of the 14.1 cDNA clone Hom-1 (9); (b) 300-bp SstI-BamHI fragment from clone 14.1, which contains homology to human IgM (probe A); (c) a 319-bp fragment containing 14.1 exon 1 sequences generated by PCR amplification using the oligonucleotides 5'-GGCCACATGGACTGGGGTGC-3' and 5'-CCACCGGCTCCTCAGGCTGG-3' (as recommended by Perkin-Elmer Corp., Norwalk, CT); (d) 200-bp SmaI fragment from clone 16.1, which shares >95% sequence similarity with exon 2 of 14.1; (e) a 60-bp fragment from nucleotide position 78-137 of GA1 exon 2 (probe D). DNA and RNA blots were hybridized overnight at 42°C in a 10% dextran sulfate, 4x SSC, 40% formamide, 0.8% Denhardt's Tris buffered solution. After hybridization, filters were washed with 2x SSC, 0.1% SDS three times at room temperature, and with 0.1x SSC, 0.1% SDS two times for 20 min at 55°C before autoradiography.

**Genomic Cloning.** A human placental DNA bacteriophage library (10° bacteriophage) or an MboI partial digest of DNA from a human pre-B cell line HPB Null inserted into Stratagene's λ DashII vector (5 x 10° bacteriophage) were screened with probes from 14.1 exon 1 or exon 3 as described previously (8, 17). Positive bacteriophage were plaque purified, and mini-lysis DNAs were prepared. Insert fragments from these clones were subcloned into Bluescript (Stratagene) for restriction mapping and DNA sequence analysis. Restriction digests were performed with indicated enzymes under conditions recommended by the manufacturer (Bethesda Research Laboratories, Gaithersburg, MD).

**Nucleotide Sequence Analysis.** The DNA sequence analysis was determined by the dideoxy chain termination method using the USB Sequenase DNA sequencing kit. Synthetic oligonucleotides (Research Genetics) were used as sequencing primers. Nucleic acid alignments and translations were done using the University of Wisconsin (Madison, WI) Sequence analysis software package (18).

## Results

**Genomic Structure of the IgM-like Gene 14.1.** Our original identification and isolation of the pre-B cell-specific 14.1 gene included a partial genomic characterization (7). With the isolation of the 14.1 cDNA, it became apparent that upstream sequences, not found on our original clone, were part of the transcription unit. A human placental DNA library was screened with a Hom-1 cDNA probe and one positive clone

---

**Figure 1.** Restriction map of human IgM locus, GA1, and 14.1 genes. A partial restriction map of the seven-gene IgM C region locus on human chromosome 22 is depicted. J and C region sequences are depicted by solid boxes and numbered. An expanded restriction map of the 14 kb EcoRI fragment containing IgM C region 1 is shown. Three coding exons of GA1 gene are depicted by solid boxes and are numbered. E=EcoRI, Bg=BglII, H=HindIII, S=Stul (note: not all Stul sites are shown). A partial restriction map of the 14.1 gene is shown. The coding exons of the 14.1 gene are depicted by solid boxes and are numbered. E=EcoRI, B=BamHI, H=HindIII, and X=XbaI.
was isolated. A partial restriction map of this clone is shown in Fig. 1. Experiments using the Hom-1 cDNA as a probe identified three areas of hybridization that define the three coding exons of the 14.1 gene. Exon 1 is positioned ~5 kb 5' of exon 2, which is in turn located ~1.2 kb 5' of exon 3. Sequence analysis of the three areas of homology to the Hom-1 cDNA proved that the clone contained the coding sequence for 14.1. These results are consistent with our prior partial characterization of the 14.1 gene as well as the recently described genomic characterization of 14.1 by Schiff et al. (15).

**Genomic Southern Analysis with the 14.1 Exon 1 Probe and Isolation of a 14.1-related Gene.** Hybridization experiments using an exon 1 probe from 14.1 on a BamHI and SstI genomic Southern blot were performed using the 14.1 exon 1 probe. This analysis identified two dominant bands at 7.6 and 3.0 kb in the BamHI digest and a 1.3- and a 1.1-kb band in the SstI digest (data not shown). Restriction mapping indicates that exon 1 of 14.1 is present on a 7.6-kb BamHI fragment and a 1.3-kb SstI fragment (Fig. 1). To clone the exon 1 sequence contained on the 3.0-kb BamHI fragment, an Mbo1 partial genomic DNA library was made from the human pre-B cell line HPB Null. Genomic Southern blot analysis showed that this gene was identical in structure in both pre-B cell and nonhematopoietic DNA (data not shown). The primary screen of the library was done with the 14.1 exon 1 probe and positive clones were counter screened with an exon 3 probe. Three clones hybridized with both probes and were plaque-purified. One of these clones contained a 7.6-kb BamHI

**Figure 2.** Nucleotide sequence of the 3 exons of the GAI and 14.1 genes. The nucleotide sequence of the GAI gene is compared to the 14.1 gene. All three exons and the predicted amino acid sequence of both genes are shown with the approximate size of introns. Introns sequences are shown in lower case letters. Numbering of exon 1 begins with the start methionine +1. Consensus polyA addition signal is indicated by underline.
fragment diagnostic of 14.1 and was not characterized further. The remaining two clones contained a 3.0-kb BamHI fragment that hybridized to the 14.1 exon 1 probe and restriction map analysis indicated that the two clones shared common sequence.

One of these clones, GL1, was analyzed in more detail and shown to contain a 14-kb EcoRI fragment. A partial restriction map of this clone was determined (Fig. 1). Comparison of the GL1 restriction map to clones containing the seven human Ig C region genes indicated that the GL1 clone was similar to the most 5' gene IgA1 (8, 19, 20). Sequence of the region homologous to IgC of clone GL1 was identical to the previously determined sequence for IgC1 region confirming the nature of the clone (Fig. 2). Sequence of the exon 1 homologous region identified a potential coding exon similar in structure to exon 1 of 14.1. This exon had one long open reading frame beginning with a start ATG capable of encoding 68 complete amino acids. This exon has a splice donor site splitting amino acid 69 that is identical to the splice donor site found at the same position of 14.1 and is a good match for a consensus splice donor site (21). This exon shows 81% and 71% homology with 14.1 exon 1 at the nucleotide and amino acid sequence level, respectively. This high degree of similarity to 14.1 exon 1 explains its identification on Southern blot and its subsequent cloning using a 14.1 exon 1 probe. Analysis of the sequence upstream of the cDNA start failed to identify a characteristic TATA or CAT box. Search of the 5' sequence failed to identify the octamer motif ATGCAAAT that has been implicated in tissue-specific expression of Ig genes (22-27).

To determine if the GL1 clone contained a sequence analogous to exon 2 of 14.1, we sequenced the area surrounding the IgA1 J region. This analysis revealed an open reading frame capable of encoding 40 amino acids, one amino acid longer than exon 2 of 14.1, with a consensus splice acceptor site at its 5' end and a consensus splice donor site at its 3' end (Fig. 2). This sequence includes the heptamer and nonamer recombination signal sequences found upstream of all J regions and in this exon would be used as coding sequence. This putative exon shares 72% and 55% similarity with the 14.1 exon 2 sequence at the nucleotide and protein level, respectively. The structure of this putative exon is such that it can join this sequence to the exon 1 and 3 sequences in an open translational reading frame.

This newly identified gene, GL1 (for germline IgA1), is capable of encoding a protein of 214 amino acids with a mol

| Exon | Amino acid | 14.1 | GL1 | λ5 |
|------|------------|------|-----|-----|
| 1    | 14.1       | 14.1 | GL1 | λ5 |
| 2    | 14.1       | 14.1 | GL1 | λ5 |
| 3    | 14.1       | 14.1 | GL1 | λ5 |

Comparison of nucleotide and amino acid sequence similarity of 14.1, GL1, and Mouse λ5; Comparison of nucleotide sequence and amino acid sequence is shown in percent identity for 14.1, GL1, and mouse λ5.
sequences, the components found in an Ig L chain codes an IgVgene-related sequence (12, 29, 30). In this com-
other pre-B cell-specific gene product, VpreB, which en-
part of a protein complex on the cell surface (10, 11). This
pre-B cell stage and precedes Ig L chain rearrangement. Re-
success to mark Bcell development (1-6). These studies have
These events, which are readily detected by Southern anal-

Discussion

Specific DNA rearrangement that juxtaposes V and J regions (and in the case of Ig H chain, diversity regions) is a prereq-
These events, which are readily detected by Southern anal-
heptamer signal sequences that are normally deleted by a VJ
quences found upstream of exon 2 as coding sequence and
the transcript would include the recombining nonamer and
region and exon 2 includes IgX1 J region sequence. While
exon structure in which exon 3 corresponds to the IgX1 C
region and exon 2 includes IgX1 J region sequence. While
this gene could be an evolutionary remnant, analysis of these
three-exons indicate that this germline gene, termed GA1,
contains several hallmarks of a functional gene. First, all three
exons are open reading frames. Second, an initiating ATG
is found in frame in the putative exon 1 sequence. Third,
consensus splice donor and acceptor sites are found at the
intron-exon junctions. Finally, a poly A addition site is found
110 bp downstream of exon 3. Transcription and processing of
these three exons would produce a transcript containing an open reading frame capable of encoding a protein with a mol wt of 23,045. This protein would use germline se-
quences found upstream of exon 2 as coding sequence and
the transcript would include the recombining nonamer and
heptamer signal sequences that are normally deleted by a V-J
rearrangement event. These results suggest that the IgX1 gene
can be expressed in two different manners. First, as we had
previously described, as a classic Ig L chain after Ig V-J gene
rearrangement (8). Second, the IgX1 gene may be expressed
in an unarranged form (GA1) utilizing the three exons related
to the 14.1 gene. Because the exon 1 and 5' portion of exon
2 are in the region deleted by V-J rearrangement, this gene
must be expressed before IgX1 V-J joining in B cell develop-
and/or be expressed in other cell types.

Based on the high degree of similarity between the pre-B
cell–specific gene 14.1 and the GA1 clone, we examined the
expression of GA1 in human pre-B cells. Our Northern anal-
ysis indicated that unlike 14.1, the GA1 clone was not ex-
pressed in pre-B cells. Of the cells examined, only an
IgX–producing B cell, DHL6, showed any GA1 transcript.
This transcript was ~800 bp in size and therefore slightly
smaller than the 14.1 transcripts seen in pre-B cells. The bio-
logical role of this germline transcript is unknown at this
time. Transcription of germline Ig genes is well documented
in both IgH and κ (for review see reference 31). Studies of these germline RNAs have shown that they can undergo RNA
processing (32). While most of these transcripts are believed to be “sterile” (incapable of producing a functional protein), at least in one case a truncated Ig H chain protein is produced. Several studies have demonstrated that germline transcription of these genes precedes gene rearrangement and may be necessary to open the chromatin structure to the rearrangement machinery. The expression of Gλ1 indicates that transcription can occur from the germline human IgA locus. This germline IgA transcript could be a forerunner of V-J gene rearrangement. Alternatively, because we have shown that there are three exons that can be spliced to produce an open reading frame, this transcript may lead to the expression of the Gλ1 sequences at the level of protein. A more extensive analysis of Gλ1 expression is being pursued to determine if transcription of the Gλ1 gene leads to production of Gλ1 protein.

Gene duplication from a single λ J and C region segments has given rise to the tandemly repeated J-C region segments of the present day seven-gene human IgA locus. This gene duplication appears to have occurred after mouse and man diverged (7, 8, 19, 20). The shared three-exon structure of the 14.1 and Gλ1 genes indicates that they arose by gene duplication from a common ancestor. Sequence comparisons of the 14.1, Gλ1, and the mouse λ genes indicate that 14.1 and Gλ1 are more similar to each other (exon 3: >89%) than to mouse λ genes (exon 3: 74% 14.1 and 75% Gλ1 to mouse λ1). This suggests that the gene duplication that gave rise to 14.1 and Gλ1 followed divergence of mouse and man. Similarly, Gλ1 is more similar to the remaining genes of the seven-gene λ locus than to 14.1 (exon 3: >95% vs. 89%), suggesting that gene duplication of the seven-gene cluster occurred after Gλ1 and 14.1 duplicated. This comparison suggests that Gλ1 may have been the original human IgA gene whose exon 2 and 3 sequences, which contain J and C regions, were duplicated to give rise to the present day human λ locus.

The conserved three-exon motif of 14.1 and Gλ1 is unlikely to have arisen recently, because the murine homologue of 14.1, mouse λ5, is organized in a similar fashion (28). This suggests that the three-exon structure found in 14.1, Gλ1, and λ5 existed in an ancestral gene before mouse-man divergence. Like man, the mouse has multiple copies of λ genes that appear to have arisen by gene duplication. A comparison of 14.1 to the four mouse λ genes and λ5 reveals that 14.1 is as similar to mouse λ 1-4 (exon 3: 72-74%) as it is to mouse λ5 (exon 3: 72%) (34). This suggests that the duplication that produced mouse λ genes 1-5 occurred from a single gene after mouse and man diverged. Based on these comparisons, we feel that the ancestral gene to 14.1/Gλ1/λ5 and the present day Ig λ genes of mouse and man is likely to have been organized in the three-exon structure seen in 14.1, Gλ1, and mouse λ5. This primordial IgA gene may have been expressed as an IgA 14.1-like protein from the germline gene or, after VJ recombination, as a canonical IgA L chain. The results described above lead to the question, is the Vpre-B-14.1 surrogate L chain complex found associated with Igμ in pre-B cells adapted from the V-J recombining Igλ genes or does it represent a primordial two-peptide Ig L chain that was later adapted to produce the single-peptide Igλ L chains? Examination of this system in more primitive immune systems may help answer this question.

We thank J. Stafford-Hollis, I. R. Kirsch, and W. M. Kuehl for critically reading the manuscript and S. Sammons for help with computer analysis.

Address correspondence to Gregory F. Hollis, Monsanto Company, AA4C 700 Chesterfield Village Parkway, St. Louis, MO 63195.

Received for publication 24 August 1990 and in revised form 19 October 1990.

References
1. Alt, F., N. Rosenberg, S. Lewis, E. Thomas, and D. Baltimore. 1981. Organization and reorganization of immunoglobulin genes in A-MuLV-transformed cells: rearrangement of heavy but not light chain genes. Cell. 27:381.
2. Korsmeyer, S.J., P.A. Hieter, J.V. Ravetch, D.G. Poplack, T.A. Waldmann, and P. Leder. 1981. Developmental hierarchy of immunoglobulin gene rearrangement in human leukemic pre-B cells. Proc. Natl. Acad. Sci. USA. 78:7096.
3. Maki, R., J. Kearney, C. Paige, and S. Tonegawa. 1982. Immunoglobulin gene rearrangements in immature B cells. Science (Wash. DC). 209:1366.
4. Perry, R., D. Kelley, C. Coleclough, and J. Kearney. 1981. Organization and expression of immunoglobulin genes in fetal liver hybridomas. Proc. Natl. Acad. Sci. USA. 78:247.
5. Reth, M., P. Ammirati, S. Jackson, and F. Alt. 1985. Related progression of a cultured pre-B cell stage. Nature (Lond.). 317:353.
6. Tonegawa, S. 1983. Somatic generation of antibody diversity. Nature (Lond.). 302:575.
7. Chang, H., E. Dmitrovsky, P. Hieter, K. Mitchell, P. Leder, L. Turoczi, I. Kirsch, and G. Hollis. 1986. Identification of three new Igλ-like genes in man. J. Exp. Med. 163:425.
8. Hieter, P., G. Hollis, S. Korsmeyer, T. Waldmann, and P. Leder. 1981. Clustered arrangements of immunoglobulin λ constant region genes in man. Nature (Lond.). 294:536.
9. Hollis, G.F., R.J. Evans, J.M. Stafford-Hollis, S.J. Korsmeyer, and J.P. McKearn. 1989. Immunoglobulin λ light chain-related genes 14.1 and 16.1 are expressed in pre-B cells and may encode the human immunoglobulin λ light chain protein. Proc. Natl. Acad. Sci. USA. 86:5552.
10. Kerr, W.G., M.D. Cooper, L. Feng, P.D. Burrows, and L.M. Hendershot. 1989. Mu heavy chains can associate with a pseudolight chain complex (\(\psi L\)) in human pre-B cell lines. *International Immunology*. 1:355.

11. Pillai, S., and D. Baltimore. 1987. Formation of disulfide-linked \(\mu 2\alpha 2\) tetramers in pre-B cells by the 18K \(\omega\)-immunoglobulin light chain. *Nature (Lond.)*. 329:172.

12. Kudo, A., and F. Melchers. 1987. A second gene, Vpre-B in the \(\lambda 5\) locus of the mouse, which appears to be selectively expressed in pre-B lymphocytes. *EMBO (Eur. Mol. Biol. Organ.) J*. 6:2267.

13. Sakaguchi, N., C. Berger, and F. Melchers. 1986. Isolation of a cDNA copy of an RNA species expressed in murine pre-B lymphocytes. *EMBO (Eur. Mol. Biol. Organ.) J*. 5:2139.

14. Sakaguchi, N., and F. Melchers. 1986. \(\lambda 5\), a new light chain-related locus selectively expressed in pre-B lymphocytes. *Nature (Lond.)*. 324:579.

15. Schiff, C., M. Bensmana, P. Guglielmi, M. Millili, M.P. Lefranc, and M. Fougereau. 1990. The immunoglobulin X-like gene cluster (14.1, 16.1, and FX1) contains genes selectively expressed in pre-B cells and is the human counterpart of the mouse \(\lambda 5\) gene. *International Immunology*. 2:201.

16. Southern, E. 1975. Detection of specific sequences among DNA fragments separated by gel electrophoresis. *J. Mol. Biol*. 98:503.

17. Gazdar, A., H. Oie, I. Kirsch, and G. Hollis. 1986. Establishment and characterization of a human plasma cell myeloma culture having a rearranged cellular myc proto-oncogene. *Blood*. 67:1542.

18. Devereux, J., P. Haeverli, and O. Smithies. 1984. A comprehensive set of sequence analysis programs for the VAX. *Nucleic Acids Res*. 12:387.

19. Dariavach, P., G. Lefranc, and M. Lefranc. 1987. Human immunoglobulin C\(\lambda 6\) gene encodes the Kern \(\psi\) \(\alpha\) \(\lambda\) chain and C\(\lambda 4\) and C\(\lambda 5\) are pseudogenes. *Proc. Natl. Acad. Sci. USA*. 84:9074.

20. Vasiceck, T.J., and P. Leder. 1990. Structure and expression of the human immunoglobulin \(\lambda\) genes. *J. Exp. Med*. 172:609.

21. Shapiro, M.B., and P. Senapathy. 1987. RNA splice junctions of different classes of eukaryotes: sequence statistics and functional implications in gene expression. *Nucleic Acids Res*. 15:7155.

22. Calame, K.L. 1985. Mechanism that regulates immunoglobulin gene expression. *Annu. Rev. Immunol*. 3:159.

23. Falkner, F.G., and H.G. Zachau. 1984. Correct transcription of an immunoglobulin \(\kappa\) gene requires an upstream fragment containing conserved sequence elements. *Nature (Lond.)*. 310:71.

24. Landolfi, N.F., J.D. Capra, and P.W. Tucker. 1986. Isolation of cell-type specific nuclear proteins with immunoglobulin V\(\kappa\) promoter region sequence. *Nature (Lond.)*. 323:548.

25. Mitchell, P.J., and R. Tjian. 1989. Transcriptional regulation in mammalian cells by sequence-specific DNA binding proteins. *Science (Wash. DC)*. 245:371.

26. Parslow, T.G., D.L. Blair, W.J. Murphy, and D.K. Granner. 1984. Structure of the 5' ends of immunoglobulin genes: a novel conserved sequence. *Proc. Natl. Acad. Sci. USA*. 74:560.

27. Wirth, T., L. Staudt, and D. Baltimore. 1987. An octamer oligonucleotide upstream of a TATA motif is sufficient for lymphoid-specific promoter activity. *Nature (Lond.)*. 329:174.

28. Kudo, A., N. Sakaguchi, and F. Melchers. 1987. Organization of the murine Ig-related \(\lambda 5\) gene transcribed selectively in pre-B lymphocytes. *EMBO (Eur. Mol. Biol. Organ.) J*. 6:103.

29. Bauer, S., K. Huebner, M. Budarf, J. Finan, J. Erikson, B. Emanuel, P. Nowell, C. Croce, and F. Melchers. 1988. The human Vpre-B gene is located on chromosome 22 near a cluster of V\(\lambda 1\) gene segments. *Immunogenetics*. 28:328.

30. Bauer, S., A. Kudo, and F. Melchers. 1988. Structure and pre-B lymphocyte restricted expression of the Vpre-B gene in humans and conservation of its structure in other mammalian species. *EMBO (Eur. Mol. Biol. Organ.) J*. 7:111.

31. Blackwell, T.K., and F.W. Alt. 1989. Mechanism and developmental program of immunoglobulin gene rearrangement in mammals. *Annu. Rev. Genet*. 23:605.

32. Martin, D.J., and B.G. Van Ness. 1990. Initiation and processing of two \(\kappa\) immunoglobulin germ line transcripts in mouse B cells. *Mol. Cell. Biol*. 10:1950.

33. Reth, M.G., and F.W. Alt. 1984. Novel immunoglobulin heavy chains are produced by DJH gene segment rearrangements in lymphoid cells. *Nature (Lond.)*. 317:418.

34. Selsing, E., J. Miller, R. Wilson, and U. Storb. 1982. Evolution of mouse immunoglobulin \(\lambda\) genes. *Proc. Natl. Acad. Sci. USA*. 79:4681.