reaching receptors, since acquisition and lavage results were the same for two different odorant concentrations (2.0 and 2.5 percent). Finally, since response decrements were of relatively short duration (1 week), it seems unlikely that widespread physical damage is the cause of our experimental observations.

The inference that some carbonyl compounds form Schiff-base linkages with proteins in the course of olfactory detection offers the simplest explanation of our findings. In vitro investigations show that acetoacetic ester plus $NaBH_3CN$ covalently modifies a model protein, AAD, which binds simple ketones reversibly. Quantitative measurements obey Eq. 1 and confirm the Schiff base–forming active site as the target of chemical blockade. Other proteins that do not bind simple ketones are not thus labeled. In vivo studies demonstrate analogous effects from the same chemical blockade in the noses of tiger salamanders. Ability to detect at least two ketones is selectively impaired. Surgical experiments confirm that the sense of smell is specifically affected. In sum, these results point toward irreversible covalent modification as a technique for identifying receptor sites in the olfactory epithelium.

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References and Notes

1. M. L. Getchell and R. C. Gesteland, *Proc. Natl. Acad. Sci. U.S.A.* **69**, 1494 (1972); S. Price, in *Biochemistry of Taste and Olfaction*, R. H. Cagan and M. R. Kare, Eds. (Academic Press, New York, 1981), pp. 69–84; D. W. Criswell, F. L. McClure, R. Schafer, K. R. Brower, *Science* **210**, 425 (1980); J. C. Delaleu and A. Holley, *Neurosci. Lett.* **37**, 251 (1983); S. Price, *Chem. Senses* **8**, 341 (1984); R. Schafer, D. W. Criswell, S. P. Fracek, Jr., K. R. Brower, *ibid.* **9**, 31 (1984); R. Schafer, S. P. Fracek, Jr., D. W. Criswell, K. R. Brower, *ibid.*, p. 55.
2. J. R. Mason and T. H. Morton, *Tetrahedron* **40**, 483 (1984).
3. ———, *Physiol. Behav.* **29**, 709 (1982).
4. I. Fridovich and F. H. Westheimer, *J. Am. Chem. Soc.* **84**, 3208 (1962).
5. F. H. Westheimer, *Methods Enzymol.* **14**, 231 (1969).
6. G. H. Hammons, F. H. Westheimer, K. Nakaoka, R. Kluger, *J. Am. Chem. Soc.* **97**, 1568 (1975).
7. S. A. Benner, J. D. Rozzell, T. H. Morton, *ibid.* **103**, 993 (1981).
8. Ethyl [3-^{14}C]acetoacetate (48.6 mCi/mmol) was purchased from New England Nuclear. Radiochemical purity ≥95 percent was assayed by adding a sample to unlabeled EAA, preparing the semicarbazone, and recrystallizing to constant specific activity.
9. In a typical experiment, duplicate samples were prepared by adding 0.06 to 5 μCi of ^{14}C-labeled EAA to solutions of 0.05 to 0.06 mg of protein in a buffer of *p*H 6 to give total volumes of 0.12 to 0.13 ml. One sample was transferred to a dialysis bag, while 0.05 ml of 0.1*M* $NaBH_3CN$ was added to the other, which was then transferred to another dialysis bag. Samples were dialyzed against buffer at 4°C. For AAD, nondialyzable radioactivity in the $NaBH_3CN$-treated samples was an order of magnitude greater than in the untreated samples. For other proteins, $NaBH_3CN$-treated and -untreated samples had the same levels of nondialyzable radioactivity.
10. R. Kluger and K. Nakaoka, *Biochemistry* **13**, 910 (1974).
11. F. H. Westheimer, *Proc. Robert A. Welch Found. Conf. Chem. Res.* **15**, 7 (1971).
12. Olfactory nerve sections (ONX) and sham surgeries (SS) were performed as double-blind studies. ONX significantly diminished responding to CH and DMDS [$F(1, 44) = 126.34$, $P < 0.001$] relative to SS. Discrimination among CH, DMDS, and BuOH was at chance level ($P > 0.25$) for ONX subjects ($n = 4$).
13. At 2.5 percent vapor saturation, ONX showed a significant decrease relative to SS in responding to CH and DMDS [$F(1, 44) = 12.5$, $P < 0.001$], but the ONX animals still showed some ability to distinguish among odorants, with mean test scores over three postoperative days of 5.3, 5.4, and 1.3. Lavage with 0.5 m*M* EAA followed by 50 m*M* $NaBH_3CN$ significantly decreased SS responding to CH when compared with prelavage, postoperative performances [$F(1, 28) = 12.34$, $P < 0.001$], but had no statistically significant effect on ONX performances ($P > 0.25$).
14. MTEAA was prepared from commercial 2,2,8-trimethyl-1,3-dioxen-4-one and 2-methylthioethanol (Aldrich) plus trace acid with distillative removal of acetone. Product (boiling point, 88° to 89°C at 0.3 torr) was twice distilled before use as a lavage agent.
15. D. G. Moulton, in *Odor Quality and Chemical Structure* (ACS Symposium Series 148), H. R. Moskowitz and C. B. Warren, Eds. (American Chemical Society, Washington, D.C., 1981), pp. 211–230.
16. Portions of this work were presented at the 13th annual meeting of the Society for Neuroscience, Boston, November 1983. We thank J. V. Connors and F. H. Westheimer for the gift of a sample of AAD, W. Silver and B. S. Gelhard for assistance in performing double-blind surgeries, and C. E. Boehm for preparing figures. This work was supported by grant NS-19424 from the National Institutes of Health.

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Diagnostic Potential for Human Malignancies of Bacterially Produced HTLV-I Envelope Protein

Abstract. *Two regions of the gene for the human T-cell leukemia virus subgroup I (HTLV-I) envelope were expressed in* Escherichia coli *by use of the vector pJLA16. One corresponds to the carboxyl terminal region of the major envelope protein p46, and the other corresponds to the transmembrane protein p21E. Reactivity of the expressed protein with human serum was tested by the Western blot procedure. Each of 11 sera tested that had been shown to contain antibodies to HTLV-I or HTLV-II by an enzyme-linked immunosorbent assay recognized the bacterially synthesized envelope proteins. There was no reaction detected when 17 control sera were tested. This system will be useful for large-scale seroepidemiological surveys for HTLV-I and related human retroviruses.*

Human T-cell leukemia virus subgroup I (HTLV-I) is a retrovirus causatively linked to certain adult lymphoid malignancies, notably adult T-cell leukemia-lymphoma (ATL) (*1*). Many isolates of this virus, identified in the United States, the Caribbean basin (*2*), southwestern Japan (*3*), Israel (*2*), Europe (*4*), and Africa (*5*), have been shown to be nearly identical (*6*). Two other isolates (HTLV-II), including one from a patient with T-cell hairy cell leukemia (*7*), are related to HTLV-I but differ significantly in antigen assays and in their genomes (*8*). A third subgroup of HTLV (HTLV-III) that is associated with the acquired immune deficiency syndrome (AIDS) has been described (*9*).

Antibodies that react with HTLV-I proteins have been found in the sera of ATL patients. These antibodies recognize both the *gag* core antigens and the envelope proteins of the virus (*10*). Viral core proteins were purified (*11*), sequenced (*12*), and used extensively in immunoassays (*13*); however, progress with the more important viral envelope proteins was slow. A limiting factor, therefore, in studies of the immune response to these viruses has been the difficulty in isolating the viral envelope proteins in pure form and in quantity.

As an alternative approach, we expressed the virus envelope protein in a bacterial vector. This procedure has the advantage that only a single viral product as defined by the structure of the input DNA is made by the bacteria. HTLV-I was suitable for such an approach because the integrated proviral DNA has been cloned (*14, 15*) and sequenced (*16*). We chose to express the HTLV-I envelope by placing it into the pJLA16 derivative (*17*) of plasmid pJL6 (*18*). This plasmid contains the 13 amino terminal codons of the bacteriophage λ *c*II gene placed under the transcriptional control of the well-regulated phage λ p_L promoter. This plasmid has been used to express sequences from *myc*, *myb*, and *ras* oncogenes (*18, 19*).

Initial attempts to express the entire HTLV-I envelope were unsuccessful, possibly because this protein can interact with the bacterial cell membrane in such a way as to be toxic to the cell. Therefore, individual fragments coding for specific regions of the envelope were inserted into pJLA6 by use of polynucleotide linkers (Fig. 1). Such plasmids were introduced into *Escherichia coli* MZ1, a strain that contains a partial λ

prophage bearing the mutant cI857 temperature-sensitive repressor. At 32°C the repressor is active, and the p_L promoter on the plasmid is repressed. At 42°C the repressor is inactive and the p_L promoter is induced, allowing a high level of expression of genes under its transcriptional control. When lysogens carrying either of the two plasmids containing different portions of the HTLV-I envelope gene were grown at 32°C and induced by shifting the temperature to 42°C, prominent bands were revealed by sodium dodecyl sulfate–polyacrylamide gel electrophoresis (SDS-PAGE) that were not found in uninduced cells or in induced cells containing the pJL6 vector alone (Fig. 2). These proteins were observed in gels of isotopically labeled bacterial extracts and in gels stained for total protein. On the basis of DNA sequence data of the envelope gene fragments, the calculated molecular sizes of the pKS300 and pKS400 proteins are 12.84 and 15.88 kilodaltons (kD), respectively. These sizes include the 1.56-kD coding sequence contributed by the amino terminal codons of the λ *c*II gene. The molecular weights of both proteins determined by SDS-PAGE are consistent with those calculated for a 321-base-pair (pKS300 insert) and a 397-base-pair (pKS400 insert) coding sequence.

The HTLV-I *env* gene codes for a glycoprotein (gp61) of molecular weight 61,000 (61K) that is cleaved into the 46K exterior glycoprotein (gp46) and the 21K transmembrane protein (gp21E) (*20*). The precise site of proteolytic cleavage has been determined by locating isotopically labeled valine residues with respect to the amino terminal end of gp21 (*21*). The cleavage of the *env* gene precursor is adjacent to the residues Arg-Arg that are also next to the proteolytic cleavage sites in the bovine leukemia virus and mouse mammary tumor virus *env* precursor (*22*). Because the Bam HI site separating the inserted fragments is close to the region coding for the proteolytic cleavage site that separates gp46 from p21E, the protein from pKS300 contains sequences corresponding to the carboxyl terminal portion of gp46, and the protein from pKS400 predominantly consists of sequences from p21E.

Sera from many patients with HTLV-I–associated ATL and certain other lymphoid malignancies contain antibodies to proteins that have been shown to be the product of the viral *env* gene (*10*). In experiments to determine whether such antibodies can recognize a bacterially synthesized envelope product, a lysate of induced MZ1[pKS400] cells containing this protein was fractionated by SDS-PAGE and transferred to nitrocellulose by electrophoretic (Western) blotting. Strips containing the transferred proteins were reacted with diluted human serum, and the antigen-antibody complexes formed were detected by incubation of the strips with ^{125}I-labeled *Staphylococcus aureus* protein A and subsequent autoradiography. Prominent bands corresponding to reaction of antibody to the 15-kD bacterial envelope product developed when the serum used was from

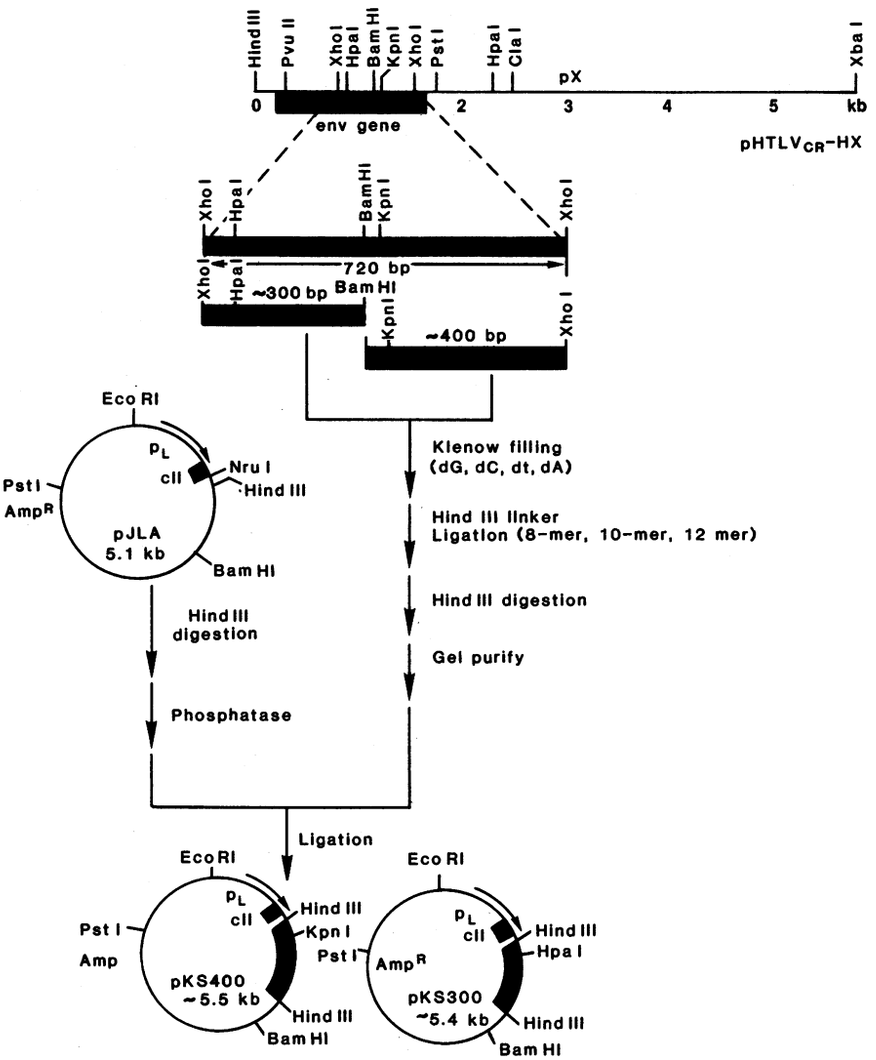

Fig. 1. Construction of plasmids pKS300 and pKS400. Plasmid pHTLV-I HX-CR was obtained by subcloning the 5.7-kb Hind III–Xba I fragment of λ CR1 (*15*) that contains envelope, pX, and long terminal repeat sequences. Recombinant DNA procedures were as described (*24*).

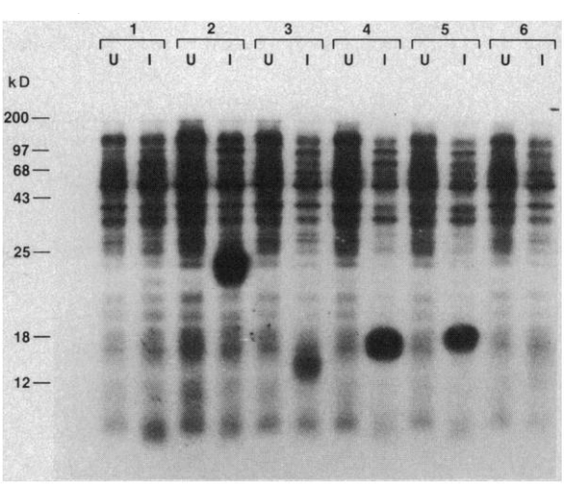

Fig. 2. Expression of the HTLV-I envelope gene in *E. coli* MZ1 cells. Cells were grown at 32°C, induced by shifting the temperature to 41°C, labeled with [^{35}S]cysteine, and lysed (*18*). Proteins were resolved by SDS-PAGE and visualized by autoradiography. Uninduced (U) and induced (I) cell extracts of expression plasmid vectors: (lane 1) pJL6 vector without insert; (lane 2) pJLcII *ras*; (lane 3) pKS300; (lane 4) pKS400.1; (lane 5) pKS400.2; (lane 6) 400-bp fragment in wrong orientation.

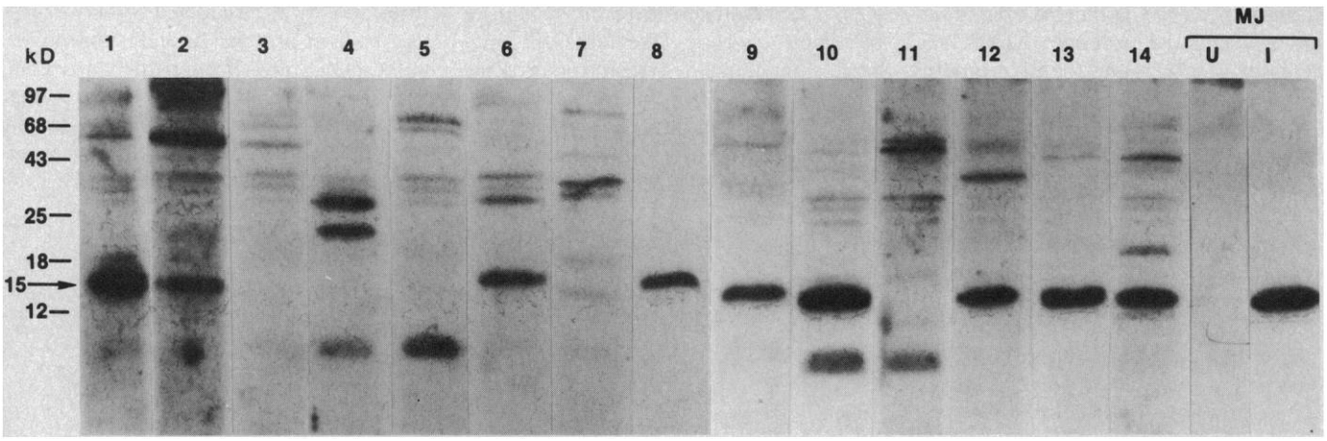

Fig. 3. Recognition of bacterially synthesized HTLV-I envelope protein by antibodies in human serum. MZ1[pKS400] cells were grown at 32°C, induced at 42°C, and lysed in the presence of 1 percent SDS and 0.1 percent β-mercaptoethanol. Protein in the extracts was resolved by SDS-PAGE and transferred to nitrocellulose by the Western blot procedure (*25*). Serum samples were from the following donors: (lane 1) American ATL patient; (lane 2) T-cell hairy cell leukemia patient Mo; (lanes 3 to 5) healthy normal donors; (lane 6) healthy relative of the ATL patient; (lane 7) healthy normal donor; (lane 8) Japanese ATL patient; (lane 9) AIDS patient found to be HTLV-II(+) by the ELISA (disrupted virus antigen); (lane 10) AIDS patient found to HTLV-I(+) by ELISA (disrupted virus antigen); (lane 11) healthy normal donor; (lane 12) American ATL patient; (lane 13) mycosis fungoides patient; (lane 14) healthy normal donor found to be HTLV-I(+) by ELISA (disrupted virus antigen). Uninduced (U) and induced (I) extracts pKS400 reacted with serum from ATL patient MJ (HTLV-I positive by ELISA). The ELISA assays were performed as described (*26*) by using HTLV-I disrupted virions.

patients with HTLV-I–associated ATL or from HTLV-I antigen (positive) individuals (Fig. 3). No such reactions were observed with sera from healthy control individuals. This procedure was used to screen a group of 28 coded sera. Antibodies that recognized the bacterially synthesized HTLV-I envelope protein sequences were found in all sera that had been shown to have antibodies to HTLV-I by an ELISA (enzyme-linked immunosorbent assay) with disrupted virions as antigen (Table 1). None of the normal control sera were found to have reacting antibodies. Antibodies from a patient (Mo) with a hairy cell leukemia (*23*), whose disease is associated with HTLV-II (*7*), strongly reacted to the protein coded for in pKS400. This indicates that there is a high degree of relatedness between the p21E region of HTLV-I and HTLV-II.

Because the bacterially synthesized HTLV-I *env* protein was recognized by antibodies in sera from an HTLV-II (positive) patient, it was of interest to see if this assay could be used to screen for HTLV-III, an even more distantly related subgroup. Therefore, we examined a number of serum samples from AIDS patients, some of whom were also seropositive for HTLV-I. The sera positive for AIDS that reacted with HTLV-I in the ELISA contained antibodies that recognized the bacterially synthesized HTLV-I *env* protein. None of the sera from AIDS patients that were HTLV-I negative contained antibodies that reacted with this protein. Because antibodies that react with HTLV-III proteins can be found in the serum of more than 90 percent of all AIDS patients (*9*), this result indicates that there is little or no cross-reaction between the carboxyl terminal portion of the envelope proteins of HTLV-I and HTLV-III. We have not completely analyzed the pKS300 protein product for reactivity in serum of leukemia patients.

These results show the importance of using bacterially synthesized proteins to study the properties of antibodies in human serum. Because the structure of the genes for such proteins can be controlled by recombinant DNA techniques, the antigens produced by these methods have a defined structure. Such antigens could be used in competition experiments to study the structure of natural antigens. The antigens we studied differ from the presumed viral envelope protein in several respects. They consist only of a small fragment of the envelope and are fused to an unrelated phage sequence at their amino terminal end. Furthermore, they may differ in secondary structure since they were synthesized in a bacterial cell and, during the immobilization process of the Western blot procedure, may have undergone denaturation. Our results show that, in spite of these factors, structures are preserved that are recognized by the antibodies to the native viral protein. Because protein can be produced in this manner in less time and for less expense, this approach should be important for large-scale epidemological surveys and for blood-bank assays.

Table 1. Antibody recognition of bacterially synthesized HTLV-I envelope in human sera. An ELISA was used to determine the presence (+) or absence (−) of antibody to HTLV-I or HTLV-II in the sera.

Donor status	Antibody	Sera tested (No.)	Sera positive (No.)
Clinically normal heterosexual	+	2	2 of 2
	−	8	0 of 8
Clinically normal homosexual	−	5	0 of 5
AIDS patients	+	2	2 of 2
	−	2	0 of 2
ATL patients	+	5	5 of 5
Mycosis fungoides patient	+	1	1 of 1
Hairy cell leukemia patient Mo	+	1	1 of 1
Lymphadenopathy syndrome patients	−	2	0 of 2

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References and Notes

1. B. J. Poiesz *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **77**, 7415 (1980); B. J. Poiesz, F. W. Ruscetti, M. S. Reitz, V. S. Kalyanaraman, R. C. Gallo, *Nature (London)* **294**, 268 (1981); R. C. Gallo *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **79**, 5680 (1982).
2. W. A. Blattner *et al.*, *Int. J. Cancer* **30**, 257 (1982); M. Popovic *et al.*, *Science* **219**, 856 (1983); R. C. Gallo *et al.*, *Cancer Res.* **43**, 3892 (1983).
3. M. Robert-Guroff *et al.*, *Science* **215**, 975 (1982); V. S. Kalyanaraman *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **79**, 1653 (1982).
4. F. A. Vyth-Dreese, P. Rumke, M. Robert-Guroff, G. de Lange, R. C. Gallo, *Int. J. Cancer* **32**, 337 (1983); D. Catovsky *et al.*, *Lancet* **1982-I**, 639 (1982); W. Blattner *et al.*, *Int J. Cancer* **30**, 257 (1982).
5. A. Fleming *et al.*, *Lancet* **1983-II**, 334 (1983); W. Saxinger *et al.*, *Science* **225**, 1473 (1984).
6. F. Wong-Staal *et al.*, *Nature (London)* **302**, 626 (1983); R. C. Gallo, in *Cancer Surveys*, J. Wyke and R. Weiss, Eds. (Oxford University Press, Oxford, 1984), p. 113.
7. V. S. Kalyanaraman *et al.*, *Science* **218**, 571 (1982).
8. G. M. Shaw *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **81**, 4544 (1984); W. A. Haseltine *et al.*, *Science* **225**, 419 (1984).
9. M. Popovic, M. G. Sarngadharan, E. Read, R. C. Gallo, *Science* **224**, 497 (1984); R. C. Gallo *et al.*, *ibid.*, p. 500; M. G. Sarngadharan *et al.*, *ibid.*, p. 506; B. Safai *et al.*, *Lancet* **1984-II**, 1438 (1984).
10. M. Essex *et al.*, *Science* **221**, 1061 (1983); P. Clapham, K. Napy, R. A. Weiss, *Proc. Natl. Aca. Sci. U.S.A.* **81** 2886 (1984).
11. V. S. Kalyanaraman, M. G. Sarngadharan, B. Poiesz, R. W. Ruscetti, R. C. Gallo, *J. Virol.* **38** 906 (1981).
12. S. Oroszlan *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **79**, 1291 (1982).
13. V. S. Kalyanaraman, M. G. Sarngadharan, P. A. Bunn, J. D. Minna, R. C. Gallo, *Nature (London)* **294**, 271 (1981).
14. M. Seiki, S. Hattori, M. Yoshida, *Proc. Natl. Acad. Sci. U.S.A.* **79**, 6899 (1982).
15. V. Manzari *et al.*, *ibid.* **80**, 1574 (1983).
16. M. Seiki, S. Hattori, Y. Hirayama, M. Yoshida, *ibid.*, p. 3618.
17. J. A. Lautenberger, A. Seth, C. Jorcyk, T. S. Papas, *Gene Anal. Techniques* **1**, 63 (1984).
18. J. A. Lautenberger, D. Court, T. S. Papas, *Gene* **23**, 75 (1983).
19. J. A. Lautenberger, L. Ulsh, T. Y. Shih, T. S. Papas, *Science* **221**, 858 (1983); J. A. Lautenberger, N. C. Kan, D. Court, T. Pry, S. Showalter, T. S. Papas, in *Gene Amplification and Analysis*, T. S. Papas, M. Rosenberg, J. Chirikjian, Eds. (Elsevier, New York, 1984), vol. 3, pp. 147–174.
20. J. Schupbach, M. G. Sarngadharan, R. C. Gallo, *Science* **224**, 607 (1984).
21. T. H. Lee, unpublished data.
22. A. M. Schultz, T. D. Copeland, S. Oroszlan, *Virology* **135**, 417 (1984).
23. A. Saxon, R. H. Stevens, D. W. Golde, *Ann. Intern. Med.* **88**, 323, 1978.
24. T. Maniatis, E. F. Fritsch, J. Sambrook, *Molecular Cloning: A Laboratory Manual* (Cold Spring Harbor Laboratory, Cold Spring Harbor, N.Y., 1982), pp. 392–397.
25. H. Towbin, T. Staehlin, G. Julian, *Proc. Natl. Acad. Sci. U.S.A.* **76**, 4350 (1979).
26. W. C. Saxinger and R. C. Gallo, *Lab. Invest.* **49**, 371 (1983).

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Cloning of the Chromosome Breakpoint of Neoplastic B Cells with the t(14;18) Chromosome Translocation

Abstract. *From an acute B-cell leukemia cell line, a DNA probe was obtained that was specific for chromosome 18 and flanked the heavy chain joining region of the immunoglobulin heavy chain locus on chromosome 14. This probe detected rearrangement of the homologous DNA segment in the leukemic cells and in follicular lymphoma cells with the t(14;18) chromosome translocation but not in other neoplastic or normal B or T cells. The probe appears to identify* bcl-2, *a gene locus on chromosome 18 (band q21) that is unrelated to known oncogenes and may be important in the pathogenesis of B-cell neoplasms with this translocation.*

Recently, we obtained a cell line, 380, derived from a 16-year-old male with acute pre–B-cell leukemia (L2 in the FAB classification) (*1*). Chromosome analysis of the leukemic cells indicated that they are pseudodiploid and carry the two chromosome translocations, t(8;14) (q24;q32) and t(14;18) (q32;q21), that are characteristic of Burkitt lymphoma (*2*) and follicular lymphoma (*3, 4*), respectively. We now describe our study with 380 cells used to clone the chromosome joining region between chromosomes 14 and 18 on one of the two $14q^+$ chromosomes resulting from these translocations.

Only the t(8;14) and t(14;18) translocations in the 380 leukemic cells, resulting in two $14q^+$ chromosomes, were detected in the karyotype (Fig. 1). The DNA's from the 380 cells were digested with Bam HI and analyzed by Southern blot transfer; the results indicated that the DNA's carry two rearranged heavy chain joining (J_H) segments of 18.5 and 14 kb (kilobases) (*1*). Since the same two DNA fragments hybridized to a C_μ (C, constant region) DNA probe (*1*), we inferred that the rearrangements occurred either within or close to the J_H segment of the two heavy chain loci. No expression of heavy chains was detected in the 380 cells (*1*), thus we also concluded that both C_μ rearrangements in these cells are unproductive. However, the t(8;14) translocation might have involved the productively rearranged μ gene leading to its activation (*1*). Deletion of both C_K genes and rearrangements of both C_λ loci were also detected in 380 cells.

A full genomic library was prepared from the DNA of the 380 cells. Genomic DNA was partially digested with the restriction enzyme Sau 3A, and DNA fragments 14 to 23 kb in length were purified by sucrose gradient centrifugation. The fragments were then ligated with DNA of the λ phage vector EMBL3A (*5*), which was cut with Bam HI. After packaging in vitro, 420,000 independent recombinant phages were screened with a probe specific for the J_H DNA segment (pHj) (Fig. 2) essentially as described for the cloning of the t(11;14) chromosome joining from neoplastic human B cells (*6*). Nine recombinant clones were obtained, and restriction map analysis allowed their classification into two groups that presumably represented sequences derived from the two $14q^+$ chromosomes.

To establish which of the two groups of recombinant clones contained the joining between chromosomes 8 and 14, we subcloned DNA fragments 5′ of both cloned J_H segments (p380j-2RR and p380j-9SS) (Fig. 2) that were free of repetitive sequences. These subclones were then used as probes in Southern blot hybridization of DNA from human cells and rodent × human hybrid cells containing either human chromosome 8, 14, or 18. Probe p380j-9SS hybridized to human DNA and to the DNA of a Chi-

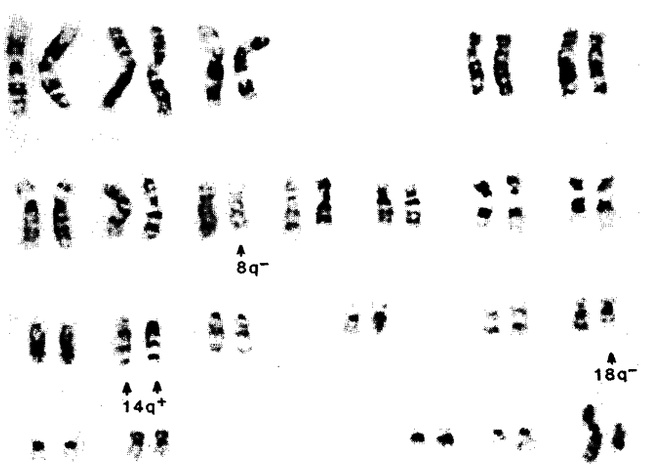

Fig. 1. Representative G-banded karyotype of the 380 cells: 46, XY, t(8;14) (q24;32), and t(14;18) (q32;q21). As a result of the two reciprocal translocations, the 380 cells have one rearranged chromosome 8 ($8q^-$), two abnormal chromosomes 14 ($14q^+$), and one abnormal chromosome 18 ($18q^-$).