JOURNAL OF VIROLOGY, June 1985, p. 889–893
0022-538X/85/060889-05$02.00/0

Vol. 54, No. 3

Nucleotide Sequence Analysis of Endogenous Murine Leukemia Virus-Related Proviral Clones Reveals Primer-Binding Sites for Glutamine tRNA

KAVE N. NIKBAKHT,[1] CHIN-YIH OU,[2] LAWRENCE R. BOONE,[2,3] PAUL L. GLOVER,[2,3] AND WEN K. YANG[2]*

University of Tennessee-Oak Ridge Graduate School of Biomedical Sciences,[1] and Biology Division, Oak Ridge National Laboratory,[2] Oak Ridge, Tennessee 37831; and National Institute of Environmental Health Sciences, Research Triangle Park, North Carolina 27612[3]

Received 19 December 1984/Accepted 25 February 1985

Nucleotide sequences of the region that corresponds to the site of tRNA primer binding for a functional retrovirus were determined in five murine leukemia virus-related sequence clones from mouse chromosomal DNA, which contain a unique 170 to 200-base-pair additional internal segment in the long terminal repeats. The 3′-terminal 18-nucleotide sequence of a major glutamine tRNA isoacceptor was found to match well with the putative primer binding site: 18 of 18 in three clones, 17 of 18 in one clone, and 16 of 18 in one clone. This implies that most of these endogenous proviral sequences of the mouse genome, if replicated as retroviruses, will be different from ecotropic murine leukemia viruses and most mammalian type C retroviruses in using glutamine tRNA, rather than proline tRNA, as a primer.

Reverse transcription of retroviral genomic RNA is initiated from a specific primer tRNA molecule, which has been identified to be tryptophan tRNA in avian sarcoma viruses and avian myeloblastosis viruses (6, 37), proline $tRNA_{1+2}$ in ecotropic murine leukemia viruses (MuLVs), feline leukemia viruses, and avian reticuloendotheliosis viruses (11, 12, 22, 34), and lysine $tRNA_3$ in murine mammary tumor viruses (23, 35). The primer function is served by the primer tRNA with its 3′-terminal, 18-nucleotide sequence (8, 13) bound to a complementary sequence located close to the 5′ end of retroviral genomic RNA (32). A detailed model of reverse transcription (10) predicts the presence of a specific tRNA binding site on the immediate 3′ side of the 5′ long terminal repeat (LTR) in the plus strand of proviral DNA; this has been generally supported by nucleotide sequence analysis of DNA clones of the aforementioned retroviruses. For example, the complementary sequence of the 3′ 18-nucleotide segment of proline $tRNA_{1+2}$ is found precisely at this site in the proviral DNA of Moloney leukemia and sarcoma viruses (7, 26, 31) as well as in the endogenous ecotropic MuLV of AKR mice, AKV (14).

The purpose of this study was to examine the tRNA primer binding site in endogenous MuLV-related sequences of laboratory mice. These MuLV-related sequences are apparently arranged in a typical provirus structure (17, 21, 25) and are more numerous than the endogenous ecotropic and xenotropic MuLV proviruses in the mouse genome (3, 9, 18, 26). They have not been isolated as infectious retrovirus, although some of the sequences may be involved in generating recombinant viruses such as mink cell cytotoxic focus-forming viruses (4, 17). It is not clear whether any of these MuLV-related proviruses possess the capacity to replicate as a retrovirus. In a previous study (21), we found that a MuLV-related proviral DNA clone from BALB/c mice contained a distinctly different tRNA primer binding site that was not complementary to the 3′-terminal sequence of proline tRNA but showed a 17-of-18 match to that of a published glutamine tRNA structure (38). Here we present experimental evidence that this glutamine tRNA shows a perfect match to the primer binding site in three of the four other MuLV-related proviruses examined.

Molecular cloning of endogenous MuLV-related sequences with Charon 9 lambda phage as the vector was performed essentially as previously described (1, 2, 21), except that *Hin*dIII-digested DNA fragments from RFM/un mice were used for insertion and an ecotropic MuLV LTR probe (19) was used for clone selection. Of the 20 clones isolated and subsequently transferred to a pBR322 vector, 12 showed positive hybridization also with a MuLV *gag-pol* sequence probe, indicating that they contained at least the 5′ half of a provirus structure. Four of these, designated pRFM-1, pRFM-9, pRFM-16, and pRFM-17, were randomly selected for further characterization.

Figure 1 shows restriction enzyme maps of the four pRFM clones (referred to below as clones 1, 9, 16, and 17) as well as a similar MuLV-related proviral clone of BALB/c mouse origin, designated AL10 (21). These five molecular clones contained various MuLV-related sequences linked to distinctly different flanking cellular sequences, indicating that they represented five individual proviruses integrated at different sites in the mouse genome. The presence of an internal *Hin*dIII site in the proviral DNA clones 1, 9, and 17 prevented the isolation of the putative 3′ portion of the proviral sequences, as did the internal *Eco*RI site in the case of AL10. The proviral sequences in these five clones all contained a few restriction enzyme sites that are also commonly found in endogenous ecotropic and xenotropic MuLVs (14, 15, 17, 24). These sites (in approximate kilobase distance from the left cell-virus junction) are located as 0.04 *Pst*I-0.5 *Kpn*I-0.5 *Sma*I sites in the LTR and 4.2 *Sal*I-4.5 *Xho*I site in the *pol* gene. In addition, apparently distinct enzyme sites common to MuLV-related proviral structures were present in most of these clones and those reported by other investigators (4, 17, 25); these include 0.4 *Bgl*II in the

* Corresponding author.

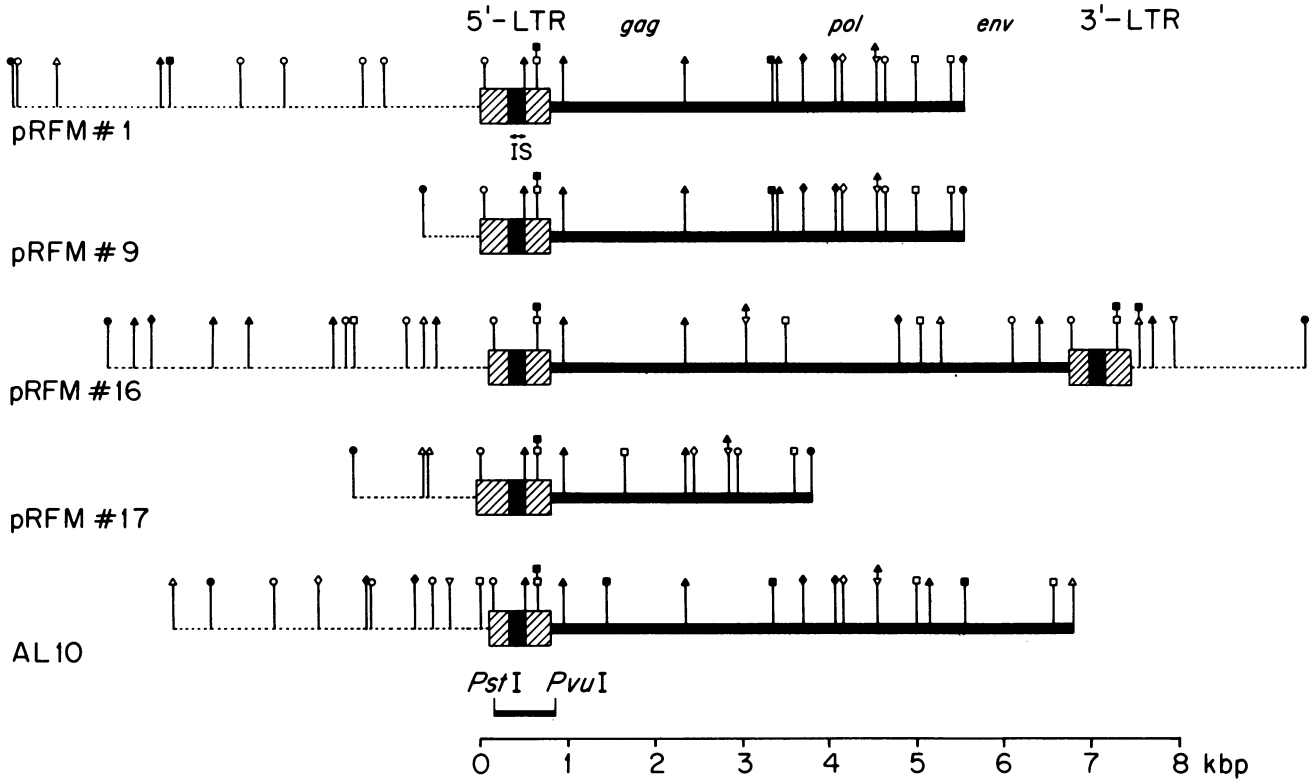

FIG. 1. Restriction endonuclease maps of five endogenous MuLV-related proviral DNA clones. pRFMs 1, 9, 16, and 17 were isolated from RFM mouse chromosomal DNA, and clone AL10 was from BALB/c mouse chromosomal DNA. Restriction sites were determined by the partial digestion technique of Smith and Birnstiel (29). The approximate sizes of the inserts were 10.9, 6.1, 13.6, 5.2, and 10.5 kbp for pRFM-1, pRFM-9, pRFM-16, pRFM-17, and AL10, respectively. The sizes of the proviral regions were 5.7 kbp (pRFM-1 and pRFM-9), 7.4 kbp (pRFM-16), 3.8 kbp (RFM-17), and 6.9 kbp (AL10). The 5' and 3' cellular flanking sequences are shown as dotted lines. The hatched area in each LTR represents the unique 170-bp sequence segment present in the U3 region. A 0.7 kbp *Pst*I-*Pvu*I fragment (shown at bottom) was of pWNB5, the molecular clone of an endogenous B-tropic ecotropic virus WN1802B derived from BALB/c mice; the LTR probe prepard from this fragment was used in the gel blots of Fig. 2 and 3. Restriction endonuclease sites are *Sma*I (□), *Kpn*I (■), *Bam*HI (◆), *Pst*I (○), *Eco*RI (△), *Xho*I (▽), *Hin*dIII (●), *Sal*I (◇), and *Bgl*II (▲).

LTR, 0.9 *Bgl*II in the *gag* gene, 3.3 *Kpn*I-3.7 *Bam*HI-4.1 *Bam*HI-4.5 *Bgl*II-5.1 *Sma*I in the *pol* gene and 6.2*Bam*HI-6.5 *Sma*I-6.8 *Eco*RI-7.9 *Bgl*II in the *env* gene. On the basis of these common and apparently distinct restriction enzyme sites, (i) clones 1 and 9 have a similar if not identical provirus, (ii) the provirus of clone 16 has a ~1.5-kilobase pair (kbp) deletion in the 5' portion of the *pol* gene, (iii) clone 17 provirus is defective at least in the *pol* gene, and (iv) clones AL10, 1 and 9 represent relatively intact proviruses (although not isolated in totality in these clones) (Fig. 1).

As demonstrated previously (17, 21), combined digestion of mouse chromosomal DNA with *Pst*I and *Kpn*I can generate three major fragments with ~600, ~550, and ~370 base pairs (bp) that are detectable by MuLV-specific LTR probe. The known ecotropic and xenotropic MuLV provirus structures (14, 15; our unpublished sequence data) are expected to produce the 370-bp *Pst*I-*Kpn*I LTR fragment. In contrast, the ~600 and the ~550-bp fragments are presumably derived from endogenous MuLV-related proviruses, as established by nucleotide sequence data showing that a novel 170 to 200-bp sequence segment is inserted within the LTR u3 region and hence makes the LTR of these proviruses larger than that of ecotropic MuLV proviruses (16, 21). Studies of the LTR size by *Pst*I-*Kpn*I digestion (Fig. 2) have demonstrated that clones 1, 9, and 17 belong to the ~600-bp class; clones 16 and AL10 belong to the 550-bp class; and the pWN41 clone of ecotropic WN1802N strain MuLV (2) belongs to the 370-bp class. Southern gel blots demonstrated that the 170 to 200-bp novel segment or homologous sequences were found to be present in the LTR u3 region of all five MuLV-related proviral clones (data not shown).

The candidate tRNA primer binding site together with its 5' flanking LTR sequence is included in a 0.6-kbp *Bgl*II fragment from clones 1, 9, 17, and AL10 and in a 1.4-kbp *Bgl*II fragment from clone 16 (Fig. 1 and 3). These fragments were therefore subcloned into pUC9 vector (27) and used for nucleotide sequence analyses in the Maxam and Gilbert procedure (20). Figure 4 shows the plus-strand DNA nucleotide sequences elucidated for the LTR u5 region and tRNA primer binding site in these five molecular clones. From these sequences, it is obvious that a *Hin*fI restriction site is present in the inverted repeat region of clone 17 but not in the same region of the other four proviral clones, and also that putative tRNA primer binding regions of clones 17 and AL10 possess a *Sau*96I site that is absent in clones 1, 9, and 16. Subsequent electrophoretic analyses could demonstrate the respective presence or absence of these specific restriction enzyme sites as predicted (data not shown), thus confirming the accuracy of the sequence data.

Three points are evident from these sequence data. First, the 3'-terminal, 18-nucleotide sequence of a rat liver major isoacceptor glutamine tRNA (38), shown at the bottom of

Fig. 4, is an 18-of-18 complementary sequence match to tRNA primer binding sites of three clones, 1, 9, and 16; and it is complementary to that of AL10 (17 of 18) and clone 17 (16 of 18). These represented the best sequence matches obtained in our search among the published tRNA structures (e.g., reference 31). Since sequences of three to four other glutamine tRNA isoacceptors present in mouse cells (39) have not been elucidated, it is not known whether these glutamine tRNA isoacceptors match better to tRNA primer binding sites of AL10 and 17 proviruses. In contrast, the 3′-terminal, 18-nucleotide sequence of proline $tRNA_{1,2}$ which serves as the tRNA primer in all ecotropic MuLV strains examined (7, 12, 14, 28, 33, 34) shows dispersed, 5- to 7-base mismatches with the primer binding sites of these five MuLV-related proviral clones. Second, when the 76-nucleotide LTR u5 sequences of the RFM mouse-derived 1, 9, 16, and 17 clones were compared with the 75-nucleotide LTR u5 sequence of the RFM mouse endogenous ecotropic MuLV (19), only 3, 7, and 8 base changes were observed in clones 1 and 9, clone 16, and clone 17, respectively. Third, the 1 or 2 bases on the immediate 3′ side of the primer binding site, designated "V" in Fig. 4, were highly variable among these proviral clones; this is similar to the reported observation of the "primer binding site-short variable region-conserved region" sequence pattern in MuLV proviruses (5).

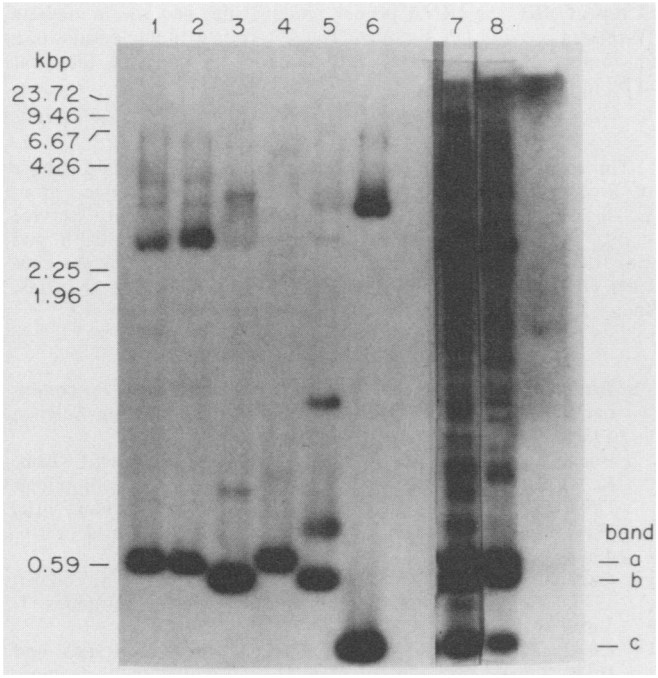

FIG. 2. Gel blot analyses of the internal *Pst*I-*Kpn*I fragments from the LTRs in endogenous MuLV-related proviral DNA clones and in mouse chromosomal DNA. Cloned DNA (0.5 μg) and chromosomal DNA (10μg) were double-digested with *Pst*I and *Kpn*I and electrophoresed at 60 V for 16 h in 1.5% agarose gel. DNA fragments were transferred to nitrocellulose filters as described by Southern (30). The LTR-specific probe (described in the legend to Fig. 1) used for hybridization had a specific activity of $\simeq 3 \times 10^8$ cpm/μg of DNA. The autoradiogram of the cloned DNAs was exposed for 1 h; the chromosomal DNAs were exposed for 24 h at −80°C. The *Pst*I-*Kpn*I internal fragments appeared as bands with high intensities (marked a, b, and c) in the low-molecular-weight region. Note that the weakly hybridizing bands seen in some lanes and a large number of high-molecular-weight DNA fragments in chromosomal DNAs were those fragments containing the 3′ 117-bp portion of the LTR. Some of the weakly hybridizing bands were not characterized. The clones examined were pRFM-1 (lane 1), pRFM-9 (lane 2), pRFM-16(LTR) subclone of pRFM-16 containing the *Bgl*II fragment with 5′-LTR (lane 3), pRFM-17 (lane 4), pAL10B3 subclone of AL10 containing the 5.1-kbp *Bam*HI fragment with 5′-LTR (lane 5), pWN41 clone of BALB/c mouse-derived N-tropic ecotropic MuLV strain WN1802N (lane 6). The chromosomal DNAs were those of RFM (lane 7) and BALB/c (lane 8) mice.

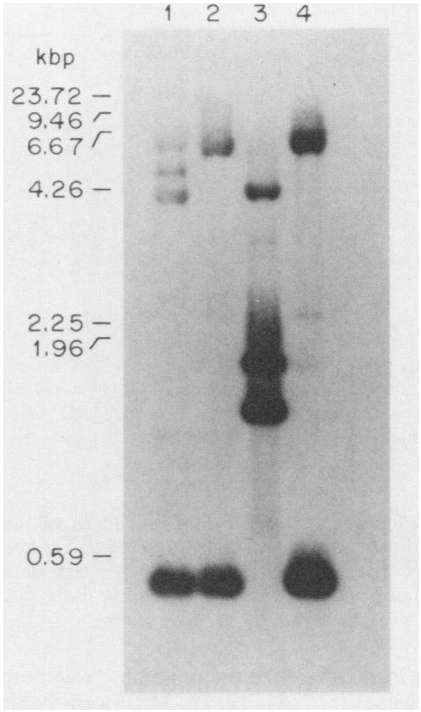

FIG. 3. Gel blot analyses of the *Bgl*II restriction endonuclease fragments from cloned endogenous MuLV-related proviral DNAs of RFM mice. A 0.5 μg sample of the cloned DNAs was digested with *Bgl*II restriction enzyme and electrophoresed at 75 V for 12 h in 0.7% agarose gel. DNA transfer and hybridization with the LTR-specific probe was described in the legend to Fig. 2. The *Bgl*II fragments containing the tRNA primer binding site appeared as 0.46-kbp, strongly-hybridizing bands in pRFM-1 (lane 1), pRFM-9 (lane 2), and pRFM-17 (lane 4), and as a 1.4-kbp band (the upper band of the two strongly hybridizing bands) in pRFM-16 (lane 3).

These results clearly demonstrate that at least three of the five randomly selected MuLV-related sequences possess a specific tRNA binding site on the immediate 3′ side of the 5′ LTR—a unique structural feature required for the tRNA-primed reverse transcription in retroviruses. This observation thus provides supportive evidence for the idea that these MuLV-related sequences were introduced into the mouse germ line through retrovirus infection and, if expressed, are capable of replication by the reverse transcription mechanism.

Since the proviral DNA clones were isolated from mouse DNA libraries by screening with an ecotropic MuLV sequence probe, and since both the LTR u5 and the *gag-pol* gene regions of these clones actually showed high degrees of sequence homology to the corresponding regions of ecotropic MuLVs (Fig. 4 and our unpublished sequence data), it was unexpected for us to find primer binding sequences for a glutamine tRNA rather than for proline $tRNA_{1+2}$. Proline tRNA has been found or implicated to be the primer for minus-strand DNA synthesis in all ecotropic MuLV strains

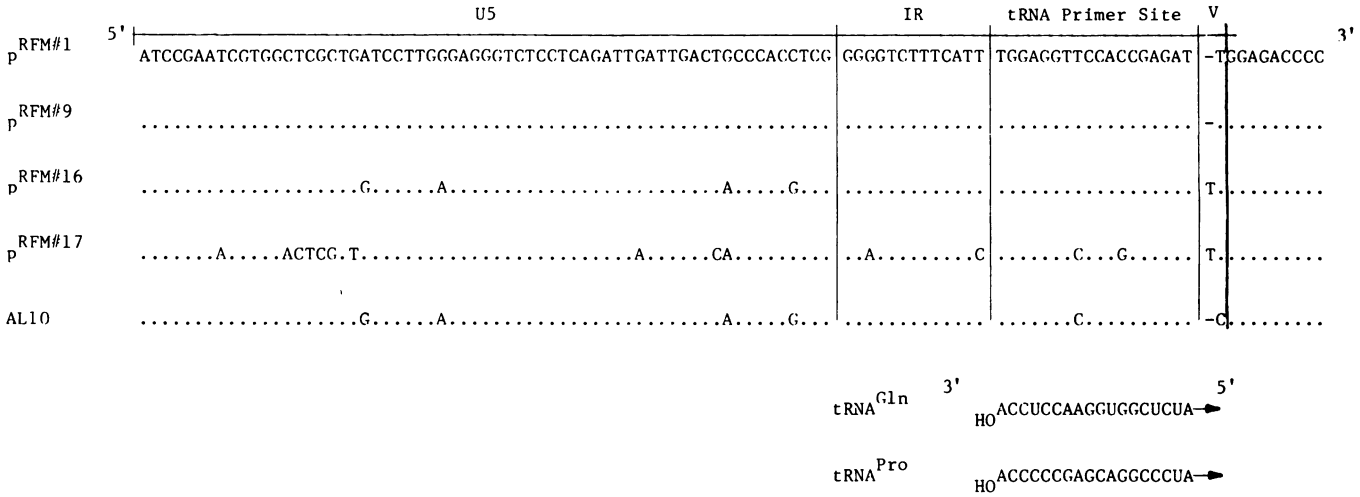

FIG. 4. Plus-stranded nucleotide sequences of the LTR u5, inverted repeat (IR), the tRNA primer binding site, and the immediate downstream variable (V) and conserved regions in five endogenous MuLV-related proviral DNA clones. The 3'-terminal 18 nucleotides of a major $tRNA^{Gln}$ isoacceptor (38) and $tRNA^{Pro}$ are shown at the bottom for comparison. With the pRFM-1 sequence as reference, identical bases of other sequences are shown as dots. Base deletions are indicated by minus signs.

as well as in many other mammalian exogenous leukemia-sarcoma viruses (5, 7, 11, 12, 22, 34, 36), probably with a few exceptions such as RD114 virus and a National Institute of Health Swiss mouse-derived xenotropic MuLV isolate (36). The present finding implicating glutamine tRNA as a primer for reverse transcription of retroviral genome is therefore novel. This characteristic primer binding site, in addition to the distinct *env* gene sequences (e.g., references 4 and 17) and the content of an extra 170 to 200-bp internal segment in the LTR u3 region (16, 21), may serve to distinguish the endogenous MuLV proviral elements of the mouse from proviruses of infectious ecotropic MuLV that show high degrees of sequence homology in most other regions of the viral genome. It is not known how the primer binding site for glutamine tRNA was acquired by this relatively abundant class of proviral sequences in the mouse germ line. In view of the marked 5 of 18 base alteration as well as the specificity of tRNA molecules involved, it is difficult to consider random point mutation as the cause for the difference between the glutamine tRNA binding site in MuLV-related proviral sequences and the proline tRNA primer binding site in ecotropic MuLV. Putative primer binding sites of AL10 and pRFM-17 proviral clones could be defective due to base mutation, but whether they could bind other glutamine tRNA isoacceptors is yet to be determined. Another possibility is that a switch from one to another tRNA primer binding occurs during the reverse transcription process. According to the commonly accepted model of reverse transcription mechanism (10), this would require either a mispaired binding of tRNA to the primer binding site of viral genomic RNA or a switch of tRNA molecule that is covalently linked to the newly synthesized minus-strand DNA. Alternatively, the ecotropic MuLVs that possess the proline tRNA primer binding site and the endogenous MuLV-related proviral sequences that possess the glutamine tRNA binding site possibly represent two separate reverse transcriptional lineages of retroviral elements that have obtained common structural gene sequences by recombination. All of these possibilities for the genetic origin of the glutamine tRNA binding site of MuLV-related proviral sequences in the mouse genome remain to be investigated.

This research has been jointly supported by the National Institute of Environmental Health Sciences (Public Health Service grant ES-40118), the National Cancer Institute (Public Health Service grants CA30308 and CA09104), and the Office of Health and Environmental Research, U.S. Department of Energy, under contract DE-AC05-840R21400 with Martin Marietta Energy Systems, Inc.

LITERATURE CITED

1. **Benton, W. D., and R. W. Davis.** 1977. Screening λ*gt* recombinant clones by hybridization to single plaques in situ. Science **196**:180–182.
2. **Boone, L. R., F. E. Myer, D. M. Yang, J. O. Kiggans, C. Koh, R. W. Tennant, and W. K. Yang.** 1983. Analysis of recombinant DNA clones of the endogenous BALB/c murine leukemia virus WN1802N: variation in long terminal repeat length. J. Virol. **45**:484–488.
3. **Callahan, R., R. E. Benveniste, M. M. Lieber, and G. J. Todaro.** 1974. Nucleic acid homology of murine type-C viral genes. J. Virol. **14**:1394–1403.
4. **Chattopadhyay, S. K., M. L. Lander, S. Gupta, E. Rands, and D. R. Lowy.** 1981. Origin of mink cytopathic focus forming (MCF) viruses: comparison with ecotropic and xenotropic murine leukemia virus genomes. Virology **113**:465–483.
5. **Chen, H. R., and W. C. Barker.** 1984. Nucleotide sequences of the retroviral long terminal repeats and their adjacent regions. Nucleic Acids Res. **12**:1767–1778.
6. **Dahlberg, J. E., R. C. Sawyer, J. M. Taylor, A. J. Faras, W. E. Levinson, H. M. Goodman, and J. M. Bishop.** 1974. Transcription of DNA from the 70*S* RNA of Rous sarcoma virus. I. Identification of a specific 4*S* RNA which serves as primer. J. Virol. **13**:1126–1133.
7. **Dhar, R., W. L. McClements, L. W. Enquist, and G. F. Vande Woude.** 1980. Nucleotide sequences of integrated Moloney sarcoma provirus long terminal repeats and their host and viral junctions. Proc. Natl. Acad. Sci. U.S.A. **77**:3937–3941.
8. **Eiden, J. J., K. Quade, and J. L. Nichols.** 1976. Interaction of tryptophan tRNA with Rous sarcoma virus 35S RNA. Nature (London) **259**:245–247.
9. **Gelb, L. D., J. B. Milstein, M. A. Martin, and S. A. Aaronson.** 1973. Characterization of murine leukemia virus-specific DNA present in normal mouse cells. Nature (London) **224**:76–79.
10. **Gilboa, E., S. W. Mitra, S. Goff, and D. Baltimore.** 1979. A detailed model of reverse transcription and tests of crucial aspects. Cell **18**:33–100.

11. **Hampe, A., M. Gobet, J. Even, C. J. Sherr, and F. Galibert.** 1983. Nucleotide sequences of feline sarcoma virus long terminal repeats and 5' leaders show extensive homology to those of other mammalian retroviruses. J. Virol. **45**:466–472.
12. **Harada, F., G. G. Peters, and J. E. Dahlberg.** 1979. The primer tRNA for Moloney murine leukemia virus DNA synthesis. Nucleotide sequence and aminoacylation of $tRNA^{Pro}$. J. Biol. Chem. **254**:10979–10985.
13. **Harada, F., R. C. Sawyer, and J. E. Dahlberg.** 1975. A primer ribonucleic acid for initiation of in vitro Rous sarcoma virus deoxyribonucleic acid synthesis. J. Biol. Chem. **250**:3487–3497.
14. **Herr, W.** 1984. Nucleotide sequence of AKV murine leukemia virus. J. Virol. **49**:471–478.
15. **Joseph, D. R.** 1981. Comparison of the restriction endonuclease maps of unintegrated proviral DNAs from four xenotropic murine leukemia viruses. J. Virol. **40**:963–970.
16. **Khan, A. S., and M. A. Martin.** 1983. Endogenous murine leukemia proviral long terminal repeats contain a unique 190-base-pair insert. Proc. Natl. Acad. Sci. U.S.A. **80**:2699–2703.
17. **Khan, A. S., W. P. Rowe, and M. A. Martin.** 1982. Cloning of endogenous murine leukemia virus-related sequences from chromosomal DNA of BALB/c and AKR/J mice: identification of an *env* progenitor of AKR-247 mink cell focus-forming proviral DNA. J. Virol. **44**:625–636.
18. **Kozak, C. A., and W. P. Rowe.** 1980. Genetic mapping of xenotropic murine leukemia virus-inducing loci in five mouse strains. J. Exp. Med. **152**:219–228.
19. **Liou, R. S., L. R. Boone, J. O. Kiggans, D. M. Yang, T. W. Wang, R. W. Tennant, and W. K. Yang.** 1983. Molecular cloning and analysis of the endogenous retrovirus chemically induced from RFM/Un mouse cell cultures. J. Virol. **46**:288–292.
20. **Maxam, A. M., and W. Gilbert.** 1977. A new method for sequencing DNA. Proc. Natl. Acad. Sci. U.S.A. **74**:560–564.
21. **Ou, C.-Y., L. R. Boone, and W. K. Yang.** 1983. A novel sequence segment and other nucleotide structural features in the long terminal repeat of a Balb/c mouse genomic leukemia virus-related DNA clone. Nucleic Acids Res. **11**:5603–5620.
22. **Peters, G. G., and C. Glover.** 1980. Low-molecular-weight RNAs and initiation of RNA-directed DNA synthesis in avian reticuloendotheliosis virus. J. Virol. **33**:708–716.
23. **Peters, G. G., and C. Glover.** 198. tRNA's and priming of RNA-directed DNA synthesis in mouse mammary tumor virus. J. Virol. **35**:31–40.
24. **Rands, E., D. R. Lowy, M. R. Lander, and S. K. Chattopadhyay.** 1981. Restriction endonuclease mapping of ecotropic murine leukemia viral DNAs: size and sequence heterogeneity of the terminal repeat sequence. Virology **108**:445–452.
25. **Roblin, R., J. M. Young, R. J. Mural, T. E. Bell, and J. N. Ihle.** 1982. Molecular cloning and characterization of murine leukemia virus-related DNA sequences from C3H/HeN mouse DNA. J. Virol. **43**:113–126.
26. **Rowe, W. P.** 1978. Leukemia viral genomes in the chromosomal DNA of the mouse. Harvey Lect. **71**:173–192.
27. **Ruther, U.** 1982. pUR250 allows rapid chemical sequencing of both DNA strands of its inserts. Nucleic Acids Res. **10**: 5765–5772.
28. **Shinnick, T. M., R. A. Lerner, and J. G. Sutcliffe.** 1981. Nucleotide sequence of Moloney murine leukemia virus. Nature (London) **293**:543–548.
29. **Smith, H. O., and M. L. Birnstiel.** 1976. A simple method for DNA restriction site mapping. Nucleic Acids Res. **3**:2387–2398.
30. **Southern, E. M.** 1975. Detection of specific sequences among DNA fragments separated by gel electrophoresis. J. Mol. Biol. **38**:503–517.
31. **Sprinzl, M., and D. H. Gauss.** 1984. Compilation of tRNA sequences. Nucleic Acids Res. **12**(Suppl.):r1–r57.
32. **Taylor, J. M., and R. Illmensee.** 1975. Site on the RNA of an avian sarcoma virus at which primer is bound. J. Virol. **16**:553–558.
33. **Van Beveren, C., F. V. Straaten, J. A. Galleshaw, and I. M. Verma.** 1981. Nucleotide sequence of the genome of a murine sarcoma virus. Cell **27**:97–108.
34. **Waters, L. C.** 1975. Transfer RNAs associated with 70S RNA of AKR murine leukemia virus. Biochem. Biophys. Res. Commun. **65**:1130–1136.
35. **Waters, L. C.** 1978. Lysine tRNA is the predominant tRNA in murine mammary tumor virus. Biochem. Biophys. Res. Commun. **81**:822–827.
36. **Waters, L. C., B. C. Mullin, E. G. Bailiff, and R. A. Popp.** 1980. Differential association of transfer RNAs with the genome of murine, feline and primate retroviruses. Biochim. Biophys. Acta **608**:112–126.
37. **Waters, L. C., B. C. Mullin, T. Ho, and W. K. Yang.** 1975. Ability of tryptophan tRNA to hybridize with 35S RNA of avian myelobastosis virus and to prime reverse transcription *in vitro*. Proc. Natl. Acad. Sci. U.S.A. **72**:2155–2159.
38. **Yang, J. A., L. W. Tai, P. F. Agris, C. W. Gehrke, and T.-W. Wong.** 1983. The nucleotide sequence of a major glutamine tRNA from rat liver. Nucleic Acids Res. **11**:1991–1996.
39. **Yang, W. K., and G. D. Novelli.** 1969. Multiple isoaccepting transfer RNA's in a mouse plasma cell tumor. Proc. Natl. Acad. Sci. U.S.A. **59**:208–215.