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Nucleotide sequence analysis of feline immunodeficiency virus: Genome organization and relationship to other lentiviruses

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ABSTRACT **We determined the complete nucleotide sequence of an infectious proviral molecular clone (FIV-14) of the feline immunodeficiency virus (FIV). FIV-14 has a genome organization similar in complexity to other lentiviruses. In addition to three large open reading frames representing the *gag*, *pol*, and *env* genes, at least four small open reading frames are present in the *pol–env* intergenic, *env*, and *env*–3′ long terminal repeat regions. Nucleotide and deduced amino acid sequence alignments of the FIV coding sequences with analogous sequences of other lentiviruses revealed significant identities only in the *gag* and *pol* genes. Phylogenetic tree analyses of *gag* and *pol* gene-encoded protein sequences demonstrate that FIV is more closely related to the ungulate lentiviruses, equine infectious anemia virus and visna virus, than to the primate lentiviruses, human and simian immunodeficiency viruses.**

Feline immunodeficiency virus (FIV) is a T-lymphotropic lentivirus that is associated with immunodeficiency and opportunistic infections in infected domestic cats (1). FIV infection occurs throughout the world and appears to be spread by horizontal transmission (2–4). Despite the ubiquitous presence of FIV, humans do not appear to be susceptible to FIV infection (1, 3).

Pathogenic lentiviral infections are well documented in other mammalian species including sheep (visna virus), goats (caprine arthritis encephalitis virus), horses (equine infectious anemia virus, EIAV), cows (bovine immunodeficiency-like virus), monkeys (simian immunodeficiency virus, SIV), and humans (human immunodeficiency virus, HIV) (5–7). These mammalian lentiviruses share similarities in genome organization, biologic properties, and the propensity for persistent infection in the natural host (8). Thus, the development of experimental models of nonhuman lentivirus infections may facilitate the design of vaccine and therapeutic strategies for HIV infection of humans.

To study molecular determinants of lentiviral pathogenesis, infectious molecular clones of proviral DNA are required. Molecular cloning of nonprimate lentiviruses rarely yields infectious proviral clones. Only one infectious clone of caprine arthritis encephalitis virus (9) and two infectious clones of bovine immunodeficiency-like virus (10) are reported, and data regarding their infectivity for animals and complete nucleotide sequences are not available. We have derived an infectious proviral DNA clone of FIV (FIV-14) and shown that progeny virions from this clone were infectious for experimentally inoculated cats (11). In this report, we describe (*i*) the complete nucleotide sequence§ and genome organization of FIV-14, (*ii*) the putative gene products of the FIV open reading frames (orfs), and (*iii*) the evolutionary relationship of FIV and other lentiviruses. Our data show that FIV is a member of the lentivirus subfamily and is distinct from characterized retroviruses.

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MATERIALS AND METHODS

Nucleotide Sequence Analysis. Proviral clone FIV-14 was subcloned [in overlapping 3- to 5-kilobase-pair (kbp) fragments] into the plasmid vectors pT7T318U (Pharmacia) and pGEM7ZF (Promega) and sequenced as described (11). Computer analyses of the nucleotide and predicted amino acid sequences were performed with the programs PC/GENE, FASTAMAIL, IFIND, and GENALIGN (IntelliGenetics, Mountain View, CA).

Primer Extension on FIV mRNA. Primer-extension analysis was performed as described by Geliebter (12) with minor modifications. Poly(A)$^+$ RNA (3 μg) from CR-FIV4 cells (11) was combined with 0.1 μg of ^{32}P-end-labeled oligonucleotide, heated to 80°C for 3 min, and annealed at 55°C for 30 min. The primer/template mixture was then incubated 30 min at 50°C in the presence of reverse transcriptase (Life Sciences, Saint Petersburg, FL) and all four dNTPs, each at 2.5 mM. The extension products were ethanol-precipitated and analyzed on a 6% polyacrylamide/urea sequencing gel.

Evolutionary Analyses. Phylogenetic trees were constructed from the distance values determined by the progressive sequence alignment method (13). The retroviral sequences used in these analyses are referenced in Doolittle *et al.* (14) except for SIVsm (SIV of sooty mangebeys, ref. 15) and SIVagm (SIV of African green monkey, ref. 16).

RESULTS AND DISCUSSION

DNA Sequence and Genome Organization. The complete nucleotide sequence of FIV-14 (9474 bp) was determined and analyzed. The genome organization of FIV is similar in complexity to other lentiviruses (Fig. 1). Three large orfs are easily identified as the major retroviral structural genes, *gag*, *pol*, and *env*. The FIV genome contains characteristic overlapping *gag* and *pol* coding sequences (Fig. 1), suggesting ribosomal frameshifting translation of the *pol* gene (17). A third large orf representing the envelope (*env*) gene is located near the 3′ end of the genome. Two small orfs (orfs 1 and 2) are located in the *pol–env* intergenic region. At least two additional small orfs (orfs 3 and 4) are found in the *env*–3′ long terminal repeat (LTR) regions (Fig. 1). Characteristics of the orfs are described below.

Abbreviations: FIV, feline immunodeficiency virus; orf, open reading frame; LTR, long terminal repeat; EIAV, equine infectious anemia virus; SIV, simian immunodeficiency virus; HIV, human immunodeficiency virus; BLV, bovine leukemia virus.

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§The sequence reported in this paper has been deposited in the GenBank database (accession no. M25381).

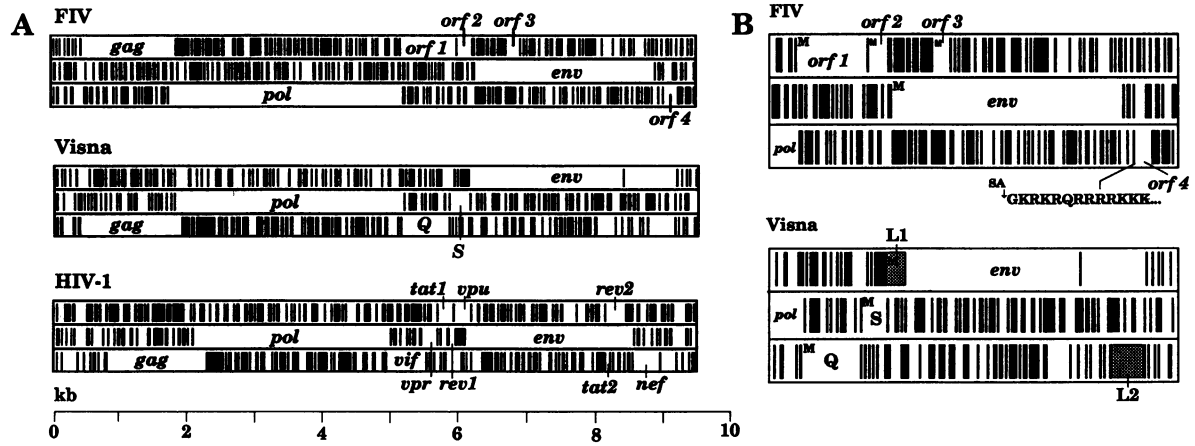

FIG. 1. (*A*) Comparison of the FIV genome organization with two other lentiviruses. The stop codon positions are indicated by the vertical lines in the three possible reading frames for each sequence. The orfs are indicated by the labeled open boxes. The sequences depicted for visna virus and HIV-1 are strains VLVCG and HXB2, respectively, and were obtained from GenBank (accession nos. M10608 and K03455). (*B*) Comparison of the small orfs of FIV and visna virus. An enlargement of the central and 3′ regions of the FIV and visna virus genomes is shown. M indicates a potential initiating methionine. The single letter amino acid code is given for the short sequence located in orf 4. SA, potential splice-acceptor site; kb, kilobases.

gag. The *gag* orf begins at base 610. A typical initiation codon (18) was located seven codons downstream. From the deduced amino acid sequence, the gag precursor is predicted to be 450 residues long (calculated molecular mass = 49.2 kDa, Table 1). By analogy to the cleavage products of the gag polyprotein precursors of EIAV and visna virus, the FIV gag proteins would be of similar size (Table 1; refs. 19 and 20). Amino acid sequence alignments of lentiviral gag proteins (data not shown) suggests that Pro-136 is the N-terminal residue of the FIV major core protein. The C-terminal residue is predicted to be Leu-362. The calculated molecular mass of this polypeptide is 25.1 kDa (Table 1) and agrees with the estimated size of the putative core protein (p28) of FIV (2, 11). Before Pro-136, the *gag* orf could encode a 135-amino acid polypeptide [gag (N terminal), Table 1]; this may correspond to the FIV-specific polypeptide (p17) observed by Western blot analysis of purified virions (2). The nucleocapsid protein is predicted by sequence alignment with EIAV and visna virus (19) to span Thr-363 to Leu-450. This 88-amino acid polypeptide (9.5 kDa, Table 1) contains two tandem Cys-His motifs that are characteristic of retroviral nucleic acid binding proteins (21).

To define highly conserved regions of *gag*, we performed a multiple alignment of lentiviral gag amino acid sequences (data not shown). The region of highest identity includes the C-terminal 81 amino acids of the putative major core protein of FIV (p28) and the N-terminal 41 amino acids of the nucleocapsid protein (p9.5). Pairwise comparisons of FIV to other lentiviruses over this region reveal significant amino acid identities: EIAV (49%), visna virus (43%), and HIV-1 (40%). The degree of relatedness observed among these viral proteins supports the previous observation that common antigenic determinants appear to be shared by the putative core proteins of FIV and the core proteins of visna virus and its closest relative, caprine arthritis encephalitis virus (11).

Table 1. Location and calculated molecular mass of FIV orfs

orf	Genome coordinates	Amino acids, no.	Molecular mass, kDa
gag	628–1978	450	49.2
gag (N terminal)*	(628–1032)	135	14.7
gag (core)*	(1033–1713)	227	25.1
gag (NCP)*	(1714–1978)	88	9.5
pol	1869–5241	1124	127.6
orf 1	5236–5989	251	29.1
orf 2	5992–6226	78	9.6
orf 3	6712–6916	68	7.7
orf 4	8955–9167	71	8.8
env (precursor)	6266–8834	856	98.1
env (outer membrane)	(6266–8098)	611	70.5
env (transmembrane)	(8099–8834)	245	27.6

Genome coordinates refer to the base locations of the first base of the initiation codon (methionine) and the first base of the stop codon in each orf with the exceptions of pol and orf 4 for which the lengths of the complete orfs are given.

**gag* gene-encoded polypeptide cleavage product and coordinates as predicted by analogy to EIAV and visna virus.

pol. The second large orf encodes polymerase gene products and overlaps the *gag* orf by 109 nucleotides. The FIV *pol* orf originates at base 1869 and potentially encodes a polypeptide of 1124 amino acids (Table 1). The predicted amino acid sequence of FIV *pol* shares significant identity with other lentiviruses. The alignment shown in Fig. 2 represents the N-terminal region of the reverse transcriptase (the region of highest identity). Pairwise comparisons of FIV to other lentiviruses reveal higher identities than observed in *gag*: visna virus (60%), HIV-1 (60%), and EIAV (61%). Alignments of the *pol* sequences before and after the reverse transcriptase region (data not shown) revealed homologies with the protease, RNase H, and endonuclease domains of the other lentiviruses (ref. 22; see the legend to Fig. 5 for the predicted boundaries). The results of these analyses (Fig. 2) for *gag* and *pol* were not unexpected since nucleotide and amino acid sequences of *gag* and *pol* have been used extensively for establishing genetic relationships among the lentiviruses (8, 19, 23).

env. The third large orf is the *env* gene based on location in the genome and structural features of the deduced amino acid sequence (Fig. 3*A*). The first methionine is 10 codons from the start of the orf and is in a preferred context for translation (18). From this initiating methionine, the *env* orf can encode a protein of 856 amino acids. To correlate calculated molecular masses (Table 1) with the observed molecular masses of the env proteins, we analyzed the viral glycoproteins in infected cells (Fig. 3*B*). Three viral-specific glycoproteins were identified: 140 kDa, designated gp140 (precursor); 100 kDa, designated gp100 (outer membrane); and 36 kDa, designated gp36 (transmembrane). The discrepancies between the observed and predicted molecular masses of the env proteins could be accounted for by 20 potential N-linked glycosylation sites present in the deduced amino acid sequence (Fig. 3*A*). When allowances for carbohydrate were

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  FIV (174) GPQIKQWPLTNEKIEALTEIVERLEREGKVKRADPNNPWNTPVFAIKK-KSGKWRMLIDFRELNKLTEKG 242
 EIAV (204) GPKIPQWPLTKEKLEGAKETVQRLLSEGKISEASDNNPYNSPIFVIKK-RSGKWRLLQDLRELNKTVQVG 272
Visna (160) GPHIAQWPLTQEKLEGLKEIVDRLEKEGKVGRAPPHWTCNTPIFCIKK-KSGKWRMLIDFRELNKQTEDL 228
  HIV (173) GPKVKQWPLTEEKIKALVEICTEMEKEGKISKIGPENPYNTPVFAIKKKDSTKWRKLVDFRELNKRTQDF 242
  SIV (253) GPKLRQWPLSKEKIIALREICEKMEKDGQLEEAPPTNPYNTPTFAIKKKDKNKWRMLIDFRELNKVTQDF 322
Consensus  GPki QWPLT EKieaL EIv rlEkEGK   A P NPyNTP FaIKK  SgKWRmLiDFRELNK Tqd

      FIV  AEVQLGLPHPAGLQIKKQVTVLDIGDAYFTIPLDPDYAPYTAFTLPRKNNAGPGRRFVWCSLPQGWILSP 312
     EIAV  TEISRGLPHPGGLIKCKHMTVLDIGDAYFTIPLDPEFRPYTAFTIPSINHQEPDKRYVWKCLPQGFVLSP 342
    Visna  AEAQLGLPHPGGLQRKKHVTILDIGDAYFTIPLYEPYRQYTCFTMLSPNNLGPCVRYYWKVLPQGWKLSP 298
      HIV  WEVQLGIPHPAGLKKKKSVTVLDVGDAYFSVPLDEDFRKYTAFTIPSINNETPGIRYQYNVLPQGWKGSP 312
      SIV  TEVQLGIPHPAGLAKRRRITVLDVGDAYFSIPLDEEFRQYTAFTLPSVNNAEPGKRYIYKVLPQGWKGSP 392
Consensus   EvQLGlPHPaGL kkK vTVLDiGDAYFtIPLDe fR YTAFT PS NN  Pg RY wkvLPQGWklSP

      FIV  LIYQSTLDNIIQPFIRQNPQLDIYQYMDDIYIGSNLSKKEHKEKVEELRKLLLWWGFETPEDKLQEEPPY 382
     EIAV  YIYQKTLQEILQPFRERYPEVQLYQYMDDLFVGSNGSKKQHKELIIELR AILQKGFETPDDKLQEVPPY 412
    Visna  AVYQFTMQKILRGWIEEHPMIQFGIYMDDIYIGSDLGLEEHRGIVNELASYIAQYGFMLPEDKRQEGYPA 368
      HIV  AIFQSSMTKILEPFRKQNPDIVIYQYMDDLYVGSDLEIGQHRTKIEELRQHLLRWGLTTPDKKHQKEPPF 382
      SIV  AIFQYTMRNVLEPFRKANPDVTLIQYMDDILIASDRTDLEHDRVVLQLKELLNGIGFSTPEEKFQKDPPF 462
Consensus  aIyQ Tm  IL PFr  nP    yQYMDDiyiGSdl   eH   v ELr  ll  GF TPedK Qe PP
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FIG. 2. Alignment of the predicted amino acid sequences of the FIV pol protein with other lentiviruses. The *pol* gene-encoded sequences represent the N-terminal region of the reverse transcriptase. Uppercase letters of the single-letter amino acid code in the consensus sequence refer to identical amino acids in at least four of the five sequences; lowercase letters refer to three matches; a blank refers to less than three. Sequences were obtained from GenBank (accession nos. M16575 and X14307) and alignments were generated by the GENALIGN program (IntelliGenetics).

included (2.1 kDa per site, ref. 25), the calculated and observed sizes of the env proteins were in good agreement.

Four main hydrophobic domains were identified (Fig. 3*A*). The first two are present in the N-terminal portion of the proposed extracellular glycoprotein (gp100). Interestingly, we did not identify a classic N-terminal signal peptide sequence. A hydrophilic segment of 94 residues precedes the first hydrophobic sequence and this, or possibly the second hydrophobic sequence, may serve as a signal peptide. This is in contrast to the env glycoproteins of EIAV, HIV-1, and SIV that have a signal sequence near the N terminus of their outer membrane glycoproteins (26–28). However, the sequence of the visna virus env is similar to FIV in that the first hydrophobic segment is located 78 amino acids from the N terminus and might correspond to the signal peptide (20). The third hydrophobic domain is probably the N terminus of the transmembrane glycoprotein (gp36). It directly follows a basic 4-amino acid peptide that is predicted to be the site of proteolytic cleavage of the env precursor molecule (gp140) into gp100 and gp36. The fourth hydrophobic segment is the predicted membrane anchor for gp36.

Significant sequence similarities between the FIV gp100 and outer membrane glycoprotein sequences of the other lentiviruses were not detected upon computer-assisted searches and alignments. Conservation of the cysteine residues and potential sites of carbohydrate addition was not detected between gp100 of FIV and the extracellular glycoproteins of visna virus, EIAV, and HIV-1. However, manual and computer-assisted comparisons of the gp36 sequence with transmembrane protein sequence alignments of visna virus, HIV-1, and EIAV (data not shown) revealed the positional conservation of two cysteine residues (Fig. 3*A*, positions 699 and 706) and four downstream potential N-linked glycosylation sites. This suggests that the FIV transmembrane protein may have functional and structural constraints in this region similar to other lentiviral transmembrane proteins.

Small orfs. Lentiviral genomes characteristically contain small orfs that encode proteins essential for the regulation of

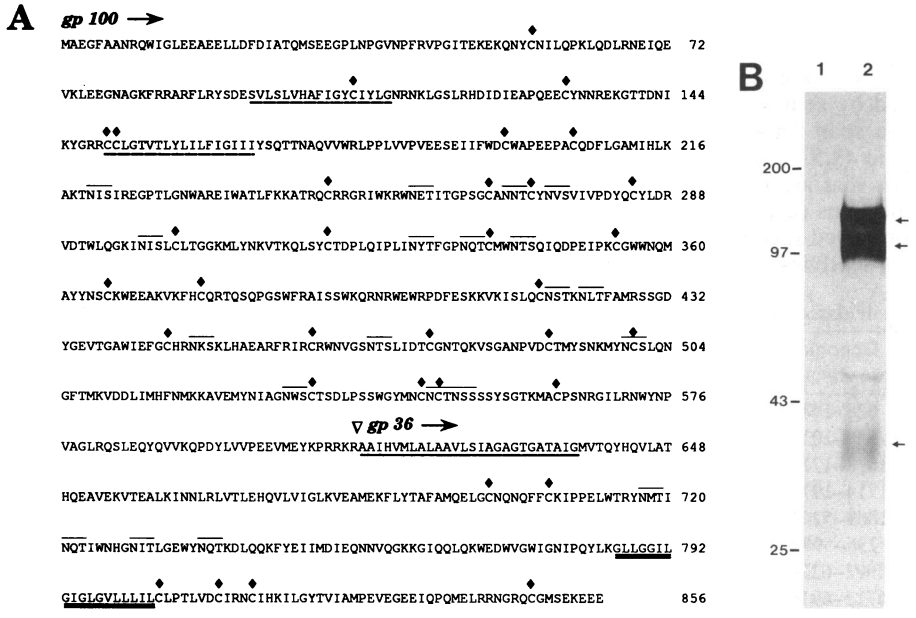

FIG. 3. Deduced amino acid sequence of the FIV-14 *env* gene. (*A*) The predicted amino acid sequence of the FIV-14 envelope glycoprotein is shown. The putative site for proteolytic cleavage of the precursor molecule gp140 is indicated by the inverted open triangle. Two hydrophobic domains in gp100 are noted by the dashed underlines. The putative N terminus and membrane-spanning domain of gp36 are indicated by the single and double underlines, respectively. Potential glycosylation sites are noted by single lines over each of the tripeptide sequences. Solid diamonds mark the locations of cysteine residues. (*B*) Radioimmunoprecipitation of FIV gp140, gp100, and gp36 from FIV-14-infected Crandell feline kidney cells. Crandell feline kidney cells persistently infected with FIV-14 were labeled for 16 hr by incubation with 100 μCi of [^{3}H]glucosamine (1 Ci = 37 GBq) per ml in RPMI medium containing one-half the normal concentration of glucose and 2% (vol/vol) fetal calf serum. Labeled polypeptides were immunoprecipitated from cell lysates with cat anti-FIV serum (lane 2) or preimmune serum (lane 1), separated on a NaDodSO$_4$/12.5% polyacrylamide gel, and visualized by fluorography as described (24). The three major glycoprotein species are indicated by the arrows. The positions of the molecular size markers (designated in kDa) are marked at the left.

viral gene expression and replication (for review, see ref. 29). At least four small orfs were identified in the FIV genome in addition to the *gag*, *pol*, and *env* orfs (Fig. 1). Attempts to align the deduced amino acid sequences of these small orfs with analogous sequences from the central and 3′ regions of the other lentiviruses were unsuccessful. However, the locations and sizes of orf 1 and orf 2 of FIV are similar to the Q and S orfs, respectively, of visna virus (Fig. 1*B*). orf 1 overlaps by 51 bases the 3′ end of the *pol* gene, has a typical initiator methionine, and can encode a polypeptide of 251 amino acids (29.1 kDa; Table 1). The Q orf is positioned similarly in the visna virus genome and its protein product has a predicted mass of 28.5 kDa (230 amino acids, ref. 23). In addition, the orf 1 gene product would be hydrophilic and basic (47% polar residues; 15% Lys + Arg), properties shared with the Q orf deduced amino acid sequence. These features are also shared by *vif* of HIV-1 (20, 30). Data describing the expression and function of Q in visna virus infection have not been reported.

orf 2 initiates with an ATG codon immediately after the stop codon of orf 1 (Fig. 1*B*). Similarly, the S orf initiates just two bases after the stop codon of Q (20). The orf 2 gene product is predicted to be approximately the same size (9.6 kDa) as the S protein (Table 1).

Based on their locations in the genome, FIV orf 3 and orf 4 may be analogous to the exons that encode the *L* gene product of visna virus (Fig. 1*B*; ref. 31). orf 3 has an initiating methionine located two codons from the start and could encode a polypeptide of 68 amino acids (Fig. 1*B*; Table 1). The FIV orf 4 deduced amino acid sequence contains a stretch of basic residues (Fig. 1*B*) that is similar to the arginine-rich domain in the second coding exon in *rev* of HIV-2, HIV-1, and SIV (15, 32, 33). Two potential splice-acceptor sites are located near the start of orf 4, indicating that this orf may serve as a 3′ exon for a spliced transcript (Fig. 1*B*).

The S orf of visna virus has been shown to encode a 10-kDa transactivator protein (31). Sequences encoding transactivating factors also have been mapped to the *pol–env* intergenic region of EIAV (34). Similar regulatory functions will likely be assigned to the central and 3′ regions in the FIV genome once the transcriptional map of FIV has been determined.

LTR. In the previously reported sequence analysis of the FIV LTR (355 bp) (11), two "TATA-like" sequences were detected, and two potential sites downstream of each sequence were proposed for initiation of RNA transcription. To determine the primary location of the RNA initiation/cap site (U3/R boundary), an oligonucleotide complementary to the primer binding site was used for primer-extension analysis on poly(A)$^+$ RNA from FIV-infected cells. Primer extension generated a major product of 162 bases that comigrated with the G nucleotide (Fig. 4), 24 bp from the second potential TATA box in the 5′ LTR, thus mapping the cap site at this nucleotide (base 217 from the start of U3). The precise site of poly(A) addition has not been determined; however, using the proposed site of the R/U5 boundary (11), the calculated lengths of the U3, R, and U5 elements of the FIV LTR are 216, 71, and 68 bp, respectively.

Evolutionary Relationship of FIV with the Other Lentiviruses. To assess the evolutionary relationship of FIV and the other members of the lentivirus family, phylogenetic trees were constructed from the amino acid sequence alignments of several lentiviral gag encoded core and nucleocapsid proteins, and the *pol*-encoded protease, reverse transcriptase, and endonuclease proteins (Fig. 5; refs. 13 and 14). In each of the four analyses, the horizontal distances and branch orders indicate that FIV is more closely related to the nonprimate lentiviruses (EIAV and visna virus) than to the primate lentiviruses. Also, these data suggest that FIV, EIAV, and visna virus diverged about the same time from a common ancestor (see below).

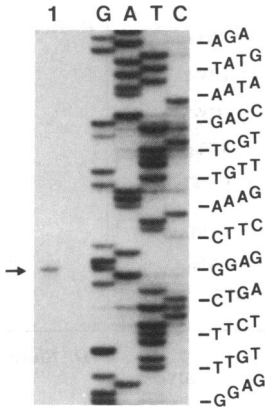

FIG. 4. Primer-extension mapping of the RNA initiation/cap site of FIV. A 21-base oligonucleotide complementary to the tRNA binding site (bases 358–378) was used for primer-extension analysis on poly(A)$^+$ RNA from FIV-infected Crandell feline kidney cells (lane 1). The same radiolabeled primer was used in dideoxynucleotide sequencing reactions on an FIV-14 plasmid subclone containing the 5′ LTR and *gag* sequences and run in parallel (lanes G, A, T, and C). The sequence depicted in the autoradiogram is given in genome sense for clarity.

Further support for the common ancestry of FIV, EIAV, and visna virus is provided by the observations of McClure *et al.* (22). Computer analyses of retroviral polymerase sequences revealed that visna virus, EIAV, and simian retrovirus type 1 contained a translocated protease-like gene segment between the RNase H and endonuclease sequences. Interestingly, these DNA sequences are not present in the *pol* gene of HIV or SIV. However, FIV contains a similar protease-like gene segment between its RNase H and endonuclease sequences (data not shown, bases 3987–4400). Moreover, the deduced amino acid sequence from this segment in FIV is ≈40% identical to the protease-like sequence in visna virus or EIAV (22). These data suggest that FIV, EIAV, and visna virus probably diverged from a common predecessor.

It is difficult to conclude whether FIV is more closely related to visna virus or EIAV from the evolutionary tree analyses due to the alternate protein-dependent tree topologies (Fig. 5). Thus, it is prudent to regard the evolutionary relationship of the FIV, visna virus, and EIAV triad as equidistant.

FIV and the Development of an Animal Model for AIDS. Our goal is the development of a small animal model to study lentiviral pathogenesis and AIDS. First, let us examine several criteria for such a model system. (*i*) The animal should be small, readily available, and easily handled. (*ii*) The pathogen should be a lentivirus with a complex genetic structure similar to primate lentiviruses (HIV and SIV). Infectious molecular clones of the virus should be available for studying molecular determinants of pathogenesis. (*iii*) Experimentally inoculated animals should become persistently infected. (*iv*) Persistent infection should lead to immunosuppression resulting in opportunistic infections. Experimental FIV infection of cats meets at least three of the four criteria. Cats are relatively inexpensive and easily handled. FIV is a lentivirus with a complex genome organization similar to primate and nonprimate lentiviruses (ref. 11 and this report). FIV establishes persistent infections in experimentally inoculated cats (ref. 2 and unpublished data).

The fourth criterion (immunosuppression and opportunistic infections) has not been a characteristic feature of experimental FIV infection (ref. 2 and unpublished data). Several features of the experimental system may partially explain these observations. To date, cats used for experimental infections have been specific-pathogen free (1, 2). Specific-pathogen-free cats do not harbor the known viral, bacterial, and parasitic pathogens carried by common domestic cats. Thus, specific-pathogen-free cats require exposure to routine feline pathogens that might cause opportunistic infections. We must consider that the only measurements of immunosuppression in cats are clinical observations; reagents for detailed study of the

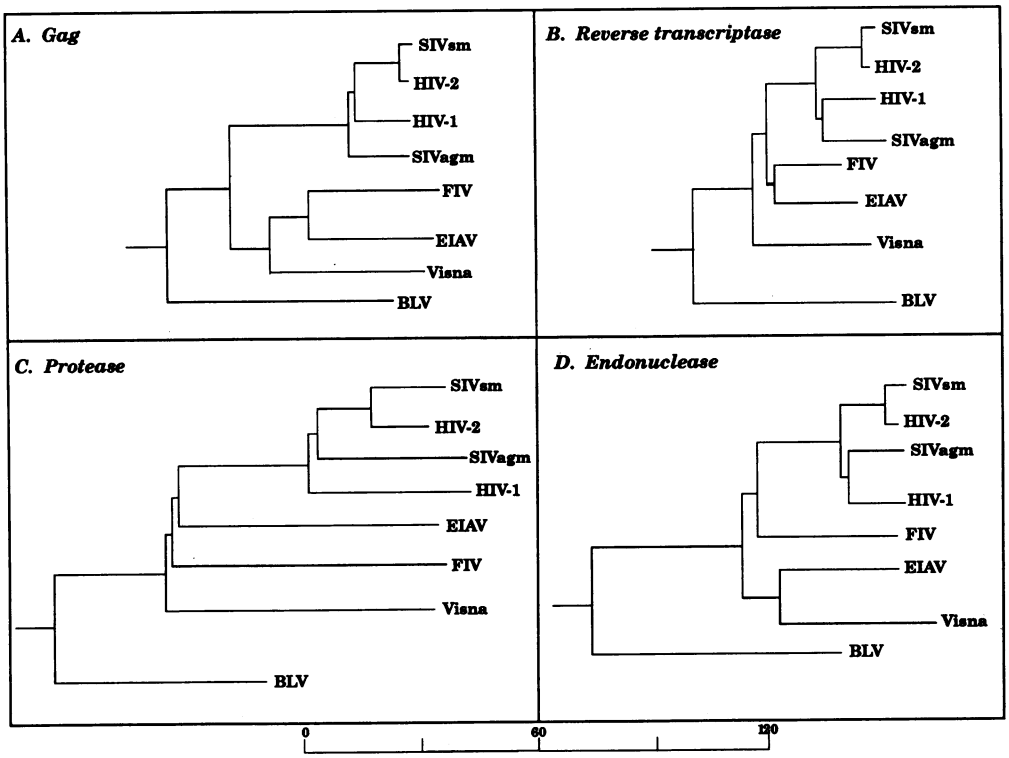

FIG. 5. Phylogenetic trees depicting the evolutionary relationship of FIV and the other lentiviruses. Trees were constructed following alignment of the gag (*A*), reverse transcriptase (*B*), protease (*C*), and endonuclease (*D*) amino acid sequences of seven lentiviruses and the type C retrovirus, BLV (bovine leukemia virus). BLV was included as the outgroup. The percent standard deviation for each tree was: gag (4.7%), reverse transcriptase (4.6%), protease (9.1%), and endonuclease (3.3%). A bar scale to obtain evolutionary distances (horizontal lengths) is included at the bottom. The length of the vertical lines is for clarity only. For reference, the amino acid and nucleotide coordinates of the FIV sequences used in the alignments were gag (Ala-1027 to Asn-1873), protease (Lys-1869 to Gln-2334), reverse transcriptase (Ile-2337 to Pro-3294), RNase H (Ile-3642 to Gln-3990, tree not shown), and endonuclease (Asp-4410 to Glu-5240).

feline immune system are not yet available. Another possibility is that FIV-induced immunosuppression may only occur after a long latency period. This might explain a previous observation that natural FIV infection has a peak incidence in older cats (3, 4). Finally, FIV may require an unidentified cofactor(s) for the induction of immunodeficiency. The molecular characterization of FIV presented in this report establishes a basis for detailed investigations of the molecular pathogenesis of FIV. Further development of the model system may require the evaluation of common domestic cats as an alternate to specific pathogen-free cats for experimental infections.

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