JOURNAL OF VIROLOGY, Aug. 1991, p. 4480–4485
0022-538X/91/084480-06$02.00/0
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Vol. 65, No. 8

Isolation of a Simian Immunodeficiency Virus Related to Human Immunodeficiency Virus Type 2 from a West African Pet Sooty Mangabey

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Received 9 October 1989/Accepted 19 April 1991

Two of 25 healthy pet sooty mangabey (SM) monkeys (*Cercocebus atys*) living in West Africa were seropositive by immunoblot when surveyed for antibody to simian immunodeficiency virus of macaques (SIVmac). SIVsmLIB1 was isolated from one of the pet sooty mangabeys. Nucleotide sequence data showed that this isolate is a member of the SIVsm/human immunodeficiency virus type 2 (HIV-2)/SIVmac group of primate lentiviruses. Furthermore, sequence comparisons revealed extensive genetic diversity among SIVsm isolates similar to that observed previously in SIV isolates from naturally infected African green monkeys. These observations provide additional evidence for monkey-human cross-species transmission of SIVsm as the source of HIV-2 infection of humans.

Simian immunodeficiency viruses (SIVs), members of the lentivirus subfamily of retroviruses, have been found so far in captive but not in wild-caught Asian macaque monkeys (mac) (members of the monkey genus *Macaca*) (1, 3) and in three species of African monkeys: (i) captive-born sooty mangabeys (sm) (*Cercocebus atys*) (6, 18, 21) in North America, (ii) East African green monkeys (agm) (*Cercopithecus aethiops*) in Kenya (5), and (iii) mandrills (mnd) (*Mandrillus sphinx*) in Gabon (25).

Serological (3, 6, 11, 18, 21) and nucleotide sequence (2, 4, 5, 9, 13) data have established that the SIVs are the closest known relatives of human immunodeficiency virus type 1 (HIV-1) and HIV-2. However, all members of the SIV group are not equally related to HIV. SIVagm and SIVmnd have genomic sequences that are about 50% identical not only to those of HIV-1 and HIV-2 but also to each other (5, 25). The SIVmac and SIVsm genomes are also about 50% homologous to the HIV-1 genome, but in contrast to SIVagm and SIVmnd, they are 75% homologous to that of HIV-2 (2, 4, 9, 13). These data indicate that SIVsm, SIVmac, and HIV-2 form a discrete subgroup of related primate lentiviruses. Identification of the natural hosts of SIVmac and SIVsm is important for understanding whether HIV-2 may have evolved from SIV in West African sooty mangabeys. This study was undertaken to determine whether sooty mangabeys in West Africa currently harbor SIV and, if so, the nature of the virus that they carry. Here we report that 2 of 25 household pet mangabeys in West Africa had antibodies to SIV and describe the characteristics of an HIV-2-related SIV isolated from one of them.

Sera were collected from 25 pet sooty mangabeys living in villages in Liberia, West Africa, in 1988 and 1989. Eleven of the pets were unwanted by their owners and were brought to the Liberian Institute for Biomedical Research (LIBR) for blood sampling. Animals were housed individually for a quarantine period of 90 days and then were housed in groups of two to three after their SIV antibody status was determined. No other monkeys were being kept at the LIBR before the initiation of the project. The LIBR maintains a colony of about 80 Liberian chimpanzees, which are housed in separate facilities and are seronegative for SIV and HIV (data not shown).

For SIV inoculation, one juvenile rhesus macaque monkey (*Macaca mulatta*) was obtained from the captive-bred colony at the California Primate Research Center (CPRC). The rhesus macaque was housed in accordance with the standards of the American Association for Accreditation of Laboratory Animal Care and, when necessary, was immobilized with 10 mg of ketamine-HCl (Parke-Davis, Morris Plains, N.J.) per kg of body weight, injected intramuscularly. Prior to use, the rhesus monkey was negative for antibodies to HIV-2, SIV, type D retrovirus, and simian T-cell leukemia virus type 1 (STLV-1), as determined by Western immunoblot (data not shown).

Sera and peripheral blood mononuclear cells (PBMCs) were prepared from pet mangabeys in Liberia, preserved in liquid nitrogen, and shipped to the University of California for in vitro analysis or inoculation into rhesus macaques as described before (20). The SIVmac strain used in this study was obtained from an infected rhesus macaque at the New England Regional Primate Research Center (3). SIV sooty mangabey California (SIVsmCALIF) was isolated from a zoo-born sooty mangabey in the United States (18). Viruses were isolated and grown in CEMx174 cells, a somatic T-cell × B-cell hybrid, as described previously (20). Either 10 or 12% polyacrylamide gels were used for immunoblots with SIV that had been purified as described before (24). Infected CEMx174 cells were prepared for thin-section electron microscopy by fixation in 2.5% glutaraldehyde and staining

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with 2% uranyl acetate as reported previously (19). FA2 and HE3 monoclonal antibodies (MAbs) are specific for the 27-kDa polypeptide of SIVmac and SIVsm, whereas HD5 reacts with a 17-kDa SIVmac polypeptide (24). The p17 MAb does not cross-react with SIVsmCALIF (24).

For genetic analysis of the *pol* region, two oligonucleotides primers were synthesized on a Cyclone DNA synthesizer (Milligen/Biosearch model 8400) for polymerase chain reaction (PCR). Two conserved oligonucleotides 1.2 kb apart were chosen as the primers. At the 5′ ends of both of the primers, a 6-base *Eco*RI recognition sequence (underlined) was added to facilitate subsequent molecular cloning of the PCR product. The sense-strand oligonucleotide was 5′GGGCGAATTCGGGAGCAATGGTGGGCGGATTACT GGC3′; the antisense-strand oligonucleotide was 5′GCGAT GAATTCTGCTGCTTCCCCTTTCC3′. These generally useful primers from conserved regions are capable of specifically amplifying *pol* gene sequences from a number of primate lentiviruses, including HIV-1HXB2, HIV-2ROD, SIVmac142, SIVsm7, and SIVagm385 (15a). A 500-ng amount of total infected-cell DNA (template) was mixed with the primers (0.2 μM each) and other reagents from the GeneAmp kit (Perkin-Elmer Cetus). The PCR conditions were as follows: denaturation at 94°C for 1 min, annealing at 42°C for 2 min, and polymerization at 72°C for 1 min, with autoextension for 5 s in each cycle. The PCR was carried out for 30 cycles under a thin layer of mineral oil. The PCR-amplified, 1.2-kbp *pol* fragment was cloned into the plasmid vector pGEM4Zf by using the *Eco*RI sites according to standard procedures. The nucleotide sequence of the purified recombinant plasmid DNA was determined by the dideoxy chain termination method (23), with Sequenase (U.S. Biochemical). The nucleotide sequence of SIVsmLIB1 was analyzed with the Pustell sequence analysis program (IBI-Pustell).

For the serological survey, plasma was tested by immunoblot for reactivity to gradient-purified SIVmac (24). Of the 25 samples tested, 2 from sooty mangabeys (SM-1 and SM-444) had antibody against SIVmac. The plasma antibodies of SM-1 (Fig. 1) and SM-444 (not shown) reacted with at least four SIV proteins: the reverse transcriptase (RT) p66, the truncated transmembrane (TM) gp32, the capsid (CA) p27, and the matrix protein (MA) p17. Plasma samples from all 25 mangabeys were seronegative for antibodies to type D retrovirus and STLV-1 (data not shown).

One of the seropositive animals, SM-1, was originally owned by two children in Robertsfield, Liberia. The children reported that it was an unwanted pet belonging to a local hunter, but beyond this information, the original owners could not be traced. The animal was a clinically healthy, sexually mature, tame female with no evidence of immunosuppressive disease after 2.5 years of observation. It was repeatedly tested over a 2-year period and remained seropositive (data not shown). Its age was estimated at 4 to 5 years at the time of the first sampling. The other seropositive pet was not available for further study, and attempts to isolate SIV from its serum have failed. Of the 23 seronegative mangabeys, 10 were brought to the LIBR over a 1- to 2-year period. They were also repeatedly tested and have remained healthy and negative for SIV antibody. Thus, 2 of 25 sooty mangabeys born in West Africa were seropositive for an SIV-like virus in this survey.

To isolate SIV from SM-1, two separate samples of PBMCs were cryopreserved in Liberia 3 months apart. The first sample was thawed at the CPRC and immediately inoculated into a captive-bred rhesus macaque seronegative

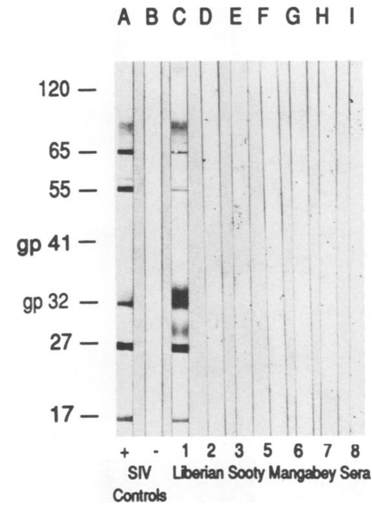

FIG. 1. Immunoblot (10% polyacrylamide gel) for SIVmac cross-reactive antibodies in serum of pet sooty mangabeys (animals 1, 2, 3, 5, 6, 7, and 8) in West Africa. A representative sample of 25 sera is shown. Two of 25 sera, SM-1 and SM-444 (not shown), were seropositive for SIVmac antibodies. Sizes are shown in kilodaltons.

for SIV, STLV-1, and type D retrovirus. Since the PBMCs from SM-1 had not been cultured in any laboratory before inoculation into the rhesus macaque, the possibility of laboratory contamination with SIV was eliminated. At 2 weeks postinoculation, rhesus monkey PBMCs were cocultivated with CEMx174 cells as described previously (20). Culture fluids were tested weekly for RT activity and were positive after 6 weeks of cocultivation. PBMCs from the infected rhesus monkey were cultured for SIV four times over an 11-month period, and all were RT positive after 4 to 8 weeks in culture (data not shown). An electron micrograph of the RT-positive culture showed a virus with typical lentiviral morphology (19) (data not shown).

For confirmation, a second sample of cryopreserved PBMCs from SM-1 was cultured with CEMx174 cells, and significant RT activity was observed after 10 weeks (data not shown). These data show that SM-1 is infected with an SIV-like virus and that this virus can infect a rhesus macaque, as previously described for SIVsm isolates from American-bred animals (21). This new West African SIV isolate was designated SIVsmLIB1 to distinguish it as the first isolate from a live sooty mangabey in West Africa.

To determine the extent of serological cross-reaction between SIVsmLIB1 and other primate lentiviruses, gradient-purified virus was reacted in an immunoblot with serum samples from HIV-1- and HIV-2-infected humans, from a Kenyan African green monkey infected with SIVagmKEN, and from SIVsmCALIF- and SIVmac-infected rhesus macaques (Fig. 2). All antisera used for comparative analysis in Fig. 2 had strong reactions with their homologous antigens, especially SU, TM, CA, MA (15), and the $p55^{gag}$ precursor (data not shown). SIVsmLIB1 cross-reacted weakly with antibody against the CA protein and the $p55^{gag}$ precursor of HIV-1. SIVagmKEN-specific serum reacted with the CA and MA proteins of SIVsmLIB1. HIV-2 antibodies reacted with the RT, CA, MA, and TM proteins of SIVsmLIB1. The strongest cross-reactions were observed with SIVmac and SIVsmCALIF antibodies, which showed cross-reactivity with CA, MA, SU, TM, and two bands in

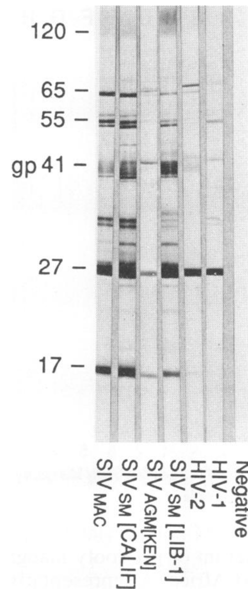

FIG. 2. Immunoblot (12% polyacrylamide gel) done with gradient-purified SIVsmLIB1 reacted with sera from the following sources; SM-1, the seropositive mangabey (note presence of the SU protein); HIV-1- and HIV-2-infected patients; an SIV-infected African green monkey; an SIVsmCALIF-infected rhesus macaque; and an SIVmac-infected rhesus macaque. Sizes are shown in kilodaltons.

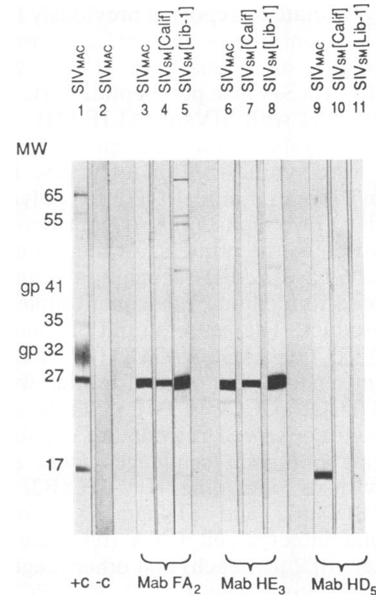

FIG. 3. Immunoblot (12% polyacrylamide gel) obtained with gradient-purified SIVmac, SIVsmCALIF, and SIVsmLIB1 and MAbs specific for SIVmac p27 (FA2 and HE3) and p17 (HD5) polypeptides. p27 MAbs reacted with all three isolates, whereas the p17 MAb reacted only with the homologous SIVmac isolate. The strain of SIV and the identity of the MAb are shown at the top and bottom, respectively, of each immunoblot. Sizes are shown in kilodaltons. +c and −c are SIVmac antibody-positive and -negative control sera, respectively.

the $p55^{gag}$ precursor region (Fig. 2). These data show a close antigenic relationship between the West African sooty mangabey virus and SIVmac and SIVsmCALIF. Note, however, that the gp120 (SU), usually the most variable structural protein, from SIVsmLIB1 reacted most strongly with plasma from the Liberian sooty mangabey, less so with the Californian sooty mangabey and SIVmac sera, and very little or not at all with sera from the other species.

SIVsmLIB1 had a nontruncated version of the TM (gp41) (Fig. 2) even though it was grown in CEMx174 cells, a human cell line. In previous studies it has been shown that growth of SIV in human cell lines can select for a TM gene with premature termination signals (14). However, the strength of the selective pressure varies with the individual SIV isolate and the cell line used.

To further characterize the antigenic relationships between SIVsmLIB1, SIVsmCALIF, and SIVmac, these viral proteins were reacted with MAbs FA2, HE3, and HD5 by immunoblot. FA2 and HE3 bind the homologous SIVmac p27, while p17 was bound by HD5 (Fig. 3) (24). SIVsmLIB1 and SIVsmCALIF had a pattern of reactivity distinct from that of SIVmac (Fig. 3). FA2 and HE3 bound the p27 protein of all three SIVs, but no reactivity was detected when the p17 MAb was reacted with either of the sooty mangabey viruses. Polyclonal serum reacted strongly with the p17 polypeptide of both SIVsmLIB1 and SIVsmCALIF, showing that fully antigenic p17 was present in the virus preparations (Fig. 2). Thus, these two SIVsm isolates were distinguishable from SIVmac by using MAbs.

For genetic analysis, 1.2 kbp of the *pol* gene, corresponding to the carboxy-terminal third of polymerase polyprotein (Pol) from SIVsmLIB1, was amplified by PCR, cloned, and sequenced. The predicted amino acid sequence and the nucleotide sequence were compared with those of the corresponding regions of the *pol* region in various isolates of HIV-1, HIV-2, SIVmac, SIVsm, and SIVagm. The results are shown in Tables 1 and 2. Clearly, SIVsm, SIVmac, and HIV-2 are more closely related to each other than to other SIV or HIV isolates. Within the SIVsm group, Pol amino acid sequence (Table 1) identity ranged from 83 to 90%, whereas SIVmac Pol had 87 to 92% amino acid sequence identity with SIVsm Pol. Pol from HIV-2 isolates had 81 to 90% amino acid sequence identity with Pol from SIVsm and 85 to 94% identity with each other. Thus, the genetic relatedness of SIVmac and HIV-2 to SIVsm is similar to the genetic relatedness among SIVsm isolates themselves. HIV-2 isolates are in some cases as close to SIVsm isolates as to other HIV-2 isolates (Table 1). Similarly, SIVsm isolates are in some cases as close to HIV-2 isolates as to other SIVsm isolates. HIV-2, SIVsm, and SIVmac are thus not distinct groups at the genetic level by these *pol* sequence analyses. Not only do SIVsm, SIVmac, and HIV-2 form a single subgroup of primate lentiviruses, but the genetic diversity of SIVsm isolates is as extensive as that observed previously for SIVagm (10, 16, 17).

The similarity between SIVsm and HIV-2 suggests cross-species transmission. What is the evidence for cross-species transmission in the direction of monkey to human rather than from human to monkey? HIV-2 and its associated disease are apparently new to the human population (7, 12) and largely confined, at least at present, to West Africa, the geographic region which is the natural habitat of sooty mangabeys and their close relatives. While SIVsm causes no apparent disease in infected sooty mangabeys (6), HIV-2 causes AIDS in humans (22). These and other features are consistent with natural infection of sooty mangabeys with

TABLE 2. Percent nucleotide identity in *pol* of primate lentiviruses[a]

Isolate	% Identical nucleotides in *pol*											
	SIVsmLIB1	SIVsmM7	SIVsmH4	SIVmac239	HIV-2ROD	HIV-2GH1	HIV-2ST	HIV-2D205	HIV-2BEN	SIVmnd	SIVagmTyol	SIVagm385
SIVsm7	83											
SIVsmH4	84	83										
SIVmac239	84	83	85									
HIV-2ROD	80	81	81	80								
HIV-2GH1	79	80	80	80	90							
HIV-2ST	80	81	81	81	91	89						
HIV-2D205	79	78	79	80	79	80	80					
HIV-2BEN	80	81	81	81	91	95	90	80				
SIVmnd	67	67	67	68	65	66	66	66	67			
SIVagmTyol	69	67	66	69	67	67	69	68	68	68		
SIVagm385	69	67	67	68	68	68	68	68	68	68	83	
HIV-1BRU	67	65	68	67	66	69	66	68	66	66	68	67

[a] The sequences used for comparison correspond to SIVsmH4 *pol* nucleotides 4055 to 5242 (HIV-1BRU *pol* nucleotides 3350 to 4538 [9, 26]). All sequences were obtained from the human retrovirus and AIDS data base at the Los Alamos National Laboratory.

TABLE 1. Percent amino acid identity in Pol of primate lentiviruses[a]

Isolate	% Identical amino acids in Pol											
	SIVsmLIB1	SIVsm7	SIVsmH4	SIVmac239	HIV-2ROD	HIV-2GH1	HIV-2ST	HIV-2D205	HIV-2BEN	SIVmnd	SIVagmTyol	SIVagm385
SIVsm7	83											
SIVsmH4	90	85										
SIVmac239	87	87	92									
HIV-2ROD	83	82	89	86								
HIV-2GH1	83	81	88	86	92							
HIV-2ST	85	84	90	89	94	91						
HIV-2D205	83	84	87	85	87	86	85					
HIV-2BEN	84	81	88	86	93	95	90	87				
SIVmnd	61	58	63	62	62	61	61	63	61			
SIVagmTyol	61	55	64	65	63	63	62	63	62	62		
SIVagm385	53	51	56	55	54	53	54	55	54	52	78	
HIV-1BRU	59	56	63	60	60	61	60	61	60	61	63	54

[a] The sequences used for comparison correspond to SIVsmH4 Pol amino acids 573 to 969 (HIV-1BRU Pol amino acids 574 to 970 [9, 26]). All sequences were obtained from the human retrovirus and AIDS data base at the Los Alamos National Laboratory.

SIVsm. We have now shown that SIVsm isolates exhibit extensive genetic diversity within the group, similar to the extensive genetic diversity demonstrated previously for SIVagm isolates from naturally infected African green monkeys (10, 16, 17). In order for this extensive variation to have been generated, SIVagm is likely to have been in the green monkey population and SIVsm in the sooty mangabey population for a prolonged period of time. Thus, the apparently recent appearance of HIV-2, the genetic similarity of SIVsm and HIV-2, the colocation of the natural habitat of sooty mangabeys and the HIV-2-endemic area, the lack of disease in sooty mangabeys and its presence in humans, and the nature of the genetic diversity together suggest but do not prove cross-species transmission from mangabeys to humans. Extensive diversity has been recently documented for HIV-2 (3a, 8a, 20a). It may be necessary to postulate either two or more separate introductions into the human population in West Africa or the presence of HIV-2 in the human population for prolonged periods sufficient to explain the HIV-2 intragroup variation.

Social issues are also relevant to the understanding of the natural history of SIV and its sooty mangabey host in West Africa. Sooty mangabeys are found primarily in Sierra Leone, Liberia, and the Ivory Coast. They are commonly used as both food and companion animals in these countries. During this study, tame mangabeys were frequently observed as family pets. Thus, ample opportunities exist for exposure of humans to SIVsm through contact with the blood, tissue, or saliva of SIV-positive animals kept as pets or killed for food. Future studies are needed to obtain SIV from free-living monkeys of all ages to establish the prevalence and genetic diversity of SIV in these wild populations.

Nucleotide sequence accession number. The SIVsmLIB1 sequence has been submitted to the Los Alamos National Laboratory AIDS data base (accession number M62651).

This research was supported by National Institutes of Health grants RR00168, RR00169, and AI-27698 to the CPRC and New England Regional Primate Research Center. Support was also provided by A125108, CA50146, and AmFAR (000591). Robert G. Webster was supported in part by Cancer Center Support (CORE) grant CA 21765 and the American Lebanese-Syrian Associated Charities.

We thank Robert Munn for electron microscopy, Robert Steen and Carole Butler for technical assistance, Sher A. Woods for administrative support, and Betty Kaufman, Sindy Staas, and Barbara Evans for secretarial services.

REFERENCES

1. **Benveniste, R. E., W. R. Morton, E. A. Clark, C.-C. Tsai, H. D. Ochs, J. M. Ward, L. Kuller, W. B. Knott, R. W. Hill, M. J. Gale, and M. E. Thouless.** 1988. Inoculation of baboons and macaques with simian immunodeficiency virus/Mne, a primate lentivirus closely related to human immunodeficiency virus type 2. J. Virol. **62:**2091–2101.
2. **Chakrabarti, L., M. Guyader, M. Alizon, M. D. Daniel, R. C. Desrosiers, P. Tiollais, and P. Sonigo.** 1987. Sequence of simian immunodeficiency virus from macaques and its relationship to other human and simian retroviruses. Nature (London) **328:**543–547.
3. **Daniel, M. D., N. L. Letvin, N. W. King, Jr., M. Kannagi, P. K. Sehgal, P. J. Kanki, M. Essex, and R. C. Desrosiers.** 1989. Isolation of T-cell tropic HTLV-III-like retrovirus from macaques. Science **228:**1201–1204.

3a. **Dietrich, U., M. Adamski, R. Kreutz, A. Seipp, H. Kühnel, and H. Rübsamen-Waigmann.** 1989. A highly divergent HIV-2-related isolate. Nature (London) **342:**948–950.

4. **Franchini, G., C. Gurgo, H. G. Guo, R. C. Gallo, E. Collalti, K. A. Fargnoli, L. F. Hall, F. Wong-Staal, and M. S. Reitz, Jr.** 1987. Sequence of simian immunodeficiency virus and its relationship to the human immunodeficiency viruses. Nature (London) **328:**539–543.
5. **Fukasawa, M., T. Miura, A. Hasegawa, S. Morikawa, H. Tsujimoto, K. Miki, T. Kitamura, and M. Hayami.** 1988. Sequence of simian immunodeficiency virus from African green monkey, a new member of the HIV-SIV group. Nature (London) **333:**457–461.
6. **Fultz, P. N., H. M. McClure, D. C. Anderson, R. B. Swenson, R. Anand, and A. Srinivasan.** 1986. Isolation of a T-lymphotropic retrovirus from naturally infected sooty mangabey monkeys (*Cercocebus atys*). Proc. Natl. Acad. Sci. USA **83:**5286–5290.
7. **Gody, M., S. A. Ouattara, and G. de The.** 1988. Clinical experience of AIDS in relation to HIV-1 and HIV-2 infection in a rural hospital in Ivory Coast, West Africa. AIDS **2:**433–436.
8. **Guyader, M., M. Emerman, P. Sonigo, F. Clavel, L. Montagnier, and M. Alizon.** 1987. Genome organization and transactivation of the human immunodeficiency virus type 2. Nature (London) **326:**662–669.

8a. **Hahn, B.** Personal communication.

9. **Hirsch, V. M., R. A. Olmsted, M. Murphey-Corb, R. H. Purcell, and P. R. Johnson.** 1989. An African primate lentivirus (SIV_{SM}) closely related to HIV-2. Nature (London) **339:**389–392.
10. **Johnson, P. R., A. Fomsgaard, J. Allan, M. Gravell, W. T. London, R. A. Olmsted, and V. M. Hirsch.** 1990. Simian immunodeficiency viruses from African green monkeys display unusual genetic diversity. J. Virol. **64:**1086–1092.
11. **Kanki, P. J., M. F. McLane, N. W. King, Jr., N. L. Letvin, R. D. Hunt, P. Sehgal, M. D. Daniel, R. C. Desrosiers, and M. Essex.** 1985. Serologic identification and characterization of a macaque T-lymphotropic retrovirus closely related to HTLV-III. Science **228:**1199–1201.
12. **Kawamura, M., S. Yamazaki, K. Ishikawa, T. B. Kwofie, H. Tsujimoto, and M. Hayami.** 1989. HIV-2 in West Africa in 1966. Lancet **i:**385.
13. **Kestler, H. W., III, Y. Li, M. Naidu, C. V. Butler, M. F. Ochs, G. Jaenel, N. W. King, M. D. Daniel, and R. C. Desrosiers.** 1988. Comparison of simian immunodeficiency virus isolates. Nature (London) **331:**619–620.
14. **Kodama, T., D. P. Wooley, Y. M. Naidu, H. W. Kestler III, M. D. Daniel, Y. Li, and R. C. Desrosiers.** 1989. Significance of premature stop codons in *env* of simian immunodeficiency virus. J. Virol. **63:**4709–4714.
15. **Leis, J., D. Baltimore, J. M. Bishop, J. Coffin, E. Fleissner, S. P. Goff, S. Oroszlan, H. Robinson, A. M. Skalka, H. M. Temin, and V. Vogt.** 1988. Standardized and simplified nomenclature for proteins common to all retroviruses. J. Virol. **62:**1808–1809.

15a. **Li, Y., and R. Desrosiers.** Unpublished data.

16. **Li, Y., Y. M. Naidu, M. D. Daniel, and R. C. Desrosiers.** 1989. Extensive genetic variability of simian immunodeficiency virus from African green monkeys. J. Virol. **63:**1800–1802.
17. **Li, Y., Y. M. Naidu, P. Fultz, M. D. Daniel, and R. C. Desrosiers.** 1989. Genetic diversity of simian immunodeficiency virus. J. Med. Primatol. **18:**261–269.
18. **Lowenstine, L. J., N. C. Pedersen, J. Higgins, K. C. Pallis, A. Uyeda, P. A. Marx, N. W. Lerche, R. J. Munn, and M. B. Gardner.** 1986. Seroepidemiologic survey of captive Old World primates for antibodies to human and simian retroviruses, and isolation of a lentivirus from sooty mangabeys (*Cercocebus atys*). Int. J. Cancer **38:**563–575.
19. **Marx, P. A., R. J. Munn, and K. I. Joy.** 1988. Computer emulation of thin section electron microscopy predicts an envelope-associated icosadeltahedral capsid for human immunodeficiency virus. Lab. Invest. **58:**112–118.
20. **Miller, C. J., N. J. Alexander, S. Sutjipto, A. A. Lackner, A. Gettie, L. J. Lowenstine, M. Jennings, and P. A. Marx.** 1989. Genital mucosal transmission of simian immunodeficiency virus: an animal model for the heterosexual transmission of human immunodeficiency virus. J. Virol. **63:**4277–4284.

20a. **Miura, T., and M. Hayami.** Personal communication.

21. **Murphey-Corb, M., L. N. Martin, S. R. S. Rangan, G. B. Baskin, B. J. Gormus, R. H. Wolf, W. A. Andes, and M. West.** 1986. Isolation of an HTLV-III related retrovirus from

macaques with simian AIDS and its possible origin in asymptomatic mangabeys. Nature (London) **321:**435–437.
22. **Romieu, I., R. Marlink, P. Kanki, S. M'Boup, and M. Essex.** 1990. HIV-2 link to AIDS in West Africa. J. Acquired Immune Defic. Syndr. **3:**220–230.
23. **Sanger, F., S. Nicklen, and A. R. Coulson.** 1977. DNA sequencing with chain-terminating inhibitors. Proc. Natl. Acad. Sci. USA **74:**5463–5467.
24. **Sutjipto, S., T. Kodama, J. Yee, A. Gettie, M. Jennings, R. C. Desrosiers, and P. A. Marx.** Characterization of monoclonal antibodies that distinguish SIV isolates from each other and from HIV-1 and 2. J. Gen. Virol., in press.
25. **Tsujimoto, H., R. W. Cooper, T. Kodama, M. Fukasawa, T. Miura, Y. Ohta, K. I. Ishikawa, M. Nakai, E. Frost, G. E. Roelants, J. Roffi, and M. Hayami.** 1988. Isolation and characterization of simian immunodeficiency virus from mandrills in Africa and its relationship to other human and simian immunodeficiency viruses. J. Virol. **62:**4044–4050.
26. **Wain-Hobson, S., P. Sonigo, O. Danos, S. Cole, and M. Alizon.** 1985. Nucleotide sequence of the AIDS virus, LAV. Cell **40:**9–17.