

## Sequence Note

# Phylogenetic Analysis of a New Chimpanzee Lentivirus SIV<sub>cpz-gab2</sub> from a Wild-Captured Chimpanzee from Gabon

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OF A TOTAL OF 50 chimpanzees wild-captured in different areas of the tropical rain forest in Gabon, two were found to be positive for HIV-1 antibodies.<sup>1</sup> From one chimpanzee a lentivirus, SIV<sub>cpz-gab1</sub>, was isolated. As a result of virological and molecular studies it was classified as an HIV-1-related nonhuman retrovirus.<sup>1,2</sup> Subsequently, another chimpanzee lentivirus, SIV<sub>cpz-ant1</sub>, has been isolated from a wild-captured chimpanzee from Zaïre.<sup>3</sup> Its molecular features are under study.

Here we report genetic evidence for infection of the second seropositive Gabonese chimpanzee<sup>1</sup> with an HIV-1-like lentivirus. This chimpanzee was about 2 years old when the mother was killed by hunters in the tropical rain forest of north-eastern Gabon. The animal stayed for 2 days in a village after having been shot. It was then brought to the primatology center of the Centre International de Recherches Médicales de Franceville (CIRM, Franceville, Gabon) for medical care, where it died of its wounds 1 week later. To the best of our knowledge, the animal had no history of inoculation with human blood products. From this chimpanzee, we have been unable thus far to isolate a lentivirus, but some of the primary peripheral blood mononuclear cells (PBMCs) have remained available in a frozen state. To investigate the genetic relationship to the SIV<sub>cpz-gab1</sub> isolate, proviral DNA was extracted from these primary PBMCs, and a 280-base pair (bp) fragment of the *pol* gene was amplified by a nested polymerase chain reaction (PCR).<sup>4</sup> Subsequently PCR fragments were cloned and sequenced. The newly obtained sequences were aligned with the corresponding *pol* gene fragments of strains belonging to different HIV-1 subtypes, SIV<sub>cpz-gab1</sub> and HIV-2<sub>ROD</sub>,<sup>5,6</sup> and a phylogenetic tree was generated (Fig. 1).<sup>7</sup> The resulting phylogenetic tree corresponded with the phylogenetic tree based on the full-length *pol* gene for the isolates for which this information was available. The new strain, referred to as SIV<sub>cpz-gab2</sub>, was positioned as an outlier of the HIV-1 group of viruses, containing genotype A, B, and D sequences, and was found to be geneti-

cally equidistant to the other viruses classified in the "outlier" group, group O.

The genetic distance between SIV<sub>cpz-gab1</sub> and SIV<sub>cpz-gab2</sub> is 14.1%. Genetic distances to the HIV-1 genotype A, B, and D strains are 13.7 to 16.3%, whereas distances to group O HIV-1

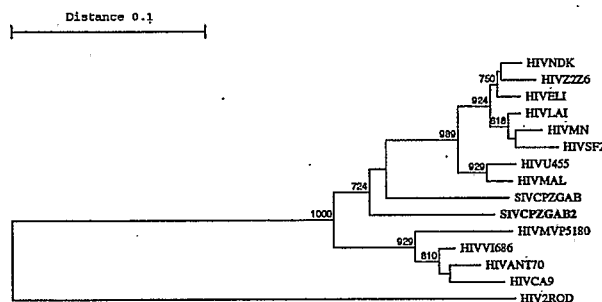


FIG. 1. Phylogenetic tree based on 280 unambiguously aligned positions of 10 HIV-1 sequences, 2 SIV<sub>cpz</sub> sequences, and 1 HIV-2 sequence. The sequence determined in this study is indicated in bold. Tree topologies were inferred by neighbor joining as previously described.<sup>7</sup> The root of the tree is placed such as to equalize its distance to the outgroup sequence HIV-2<sub>ROD</sub> and its average to the HIV-1 and SIV<sub>cpz</sub> sequences. The distance between two sequences is obtained by summing the lengths of the connecting horizontal branches, using the scale on top. The number of bootstrap trees out of 1000 replications supporting a particular phylogenetic group in more than 70% is placed alongside the node considered. Branches supported by less than 70% of the bootstrap trees are not reliable. The nucleotide sequence data of SIV<sub>cpz-gab2</sub> was deposited in the EMBL, GenBank, and DDBJ Nucleotide Sequence Databases under the following accession number U11495: All other sequences shown are available through GenBank and the HIV Sequence Database.

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	ZZ26	NDK	ELI	LAI	SF2	MN	MAL	U455	CPZGAB	CPZGAB2	MVP5180	VI686	CA9	ANT70	ROD
NDK	2.91	0.00													
ELI	3.28	2.54	0.00												
LAI	3.66	3.66	3.29	0.00											
SF2	5.94	5.17	5.56	3.66	0.00										
MN	5.17	4.41	4.79	2.17	3.66	0.00									
MAL	7.11	6.72	6.72	5.94	7.11	7.51	0.00								
U455	7.11	6.72	6.72	6.72	7.11	8.30	2.91	0.00							
CPZGAB	14.54	12.40	12.40	13.67	16.74	14.97	13.25	13.25	0.00						
CPZGAB2	16.29	15.41	14.10	14.10	16.29	15.41	14.10	13.67	14.10	0.00					
MVP5180	19.93	19.00	19.46	20.39	22.78	21.34	19.46	18.54	16.74	18.09	0.00				
VI686	17.18	14.97	14.97	15.85	18.09	18.09	15.41	14.10	15.41	15.41	7.90	0.00			
CA9	21.34	18.09	19.00	19.93	20.39	20.39	19.93	18.54	17.63	15.85	9.51	4.79	0.00		
ANT70	19.54	17.25	17.25	18.16	20.48	20.48	17.70	16.36	15.03	15.47	7.53	2.55	4.43	0.00	
ROD	54.90	53.43	51.99	51.99	52.70	53.43	52.70	52.70	52.70	51.27	49.87	53.43	50.57	52.25	0.00

FIG. 2. Similarity distance matrix showing pairwise comparisons of nucleotide sequences of a 280-bp fragment in the *pol* gene.<sup>4</sup> The numbers shown indicate the percentage of positions that differ between two nucleotide sequences.

strains are 15.4 to 18.5% (Fig. 2). As apparent from the phylogenetic tree, SIV<sub>cpz-gab</sub> and SIV<sub>cpz-gab2</sub> strains are more related to the HIV-1 strains not belonging to group O, over this relatively short stretch of sequence. SIV<sub>cpz-gab</sub> and SIV<sub>cpz-gab2</sub> cluster with the HIV-1 strains, not related to group O, in 724 of 1000 bootstrap trees (72.4%). On the basis of their respective distances to each other and to the HIV-1 strains, SIV<sub>cpz-gab</sub> and SIV<sub>cpz-gab2</sub> can be assigned as representative for two distinct genetic lineages of HIV-1-related chimpanzee lentiviruses.

In conclusion, this study supports natural infection of chimpanzees with an HIV-1-related virus in the wild.

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