

Sequence Note

Genetic Analysis of HIV Type 2 from Ghana and Guinea-Bissau, West Africa

KOICHI ISHIKAWA,^{1,2} WOUTER JANSSENS,³ JACOB S. BANOR,² TEIICHIRO SHINNO,¹
JOÃO PIEDADE,⁴ TETSUTARO SATA,¹ WILLIAM K. AMPOFO,² JAMES A.M. BRANDFUL,²
YOSHIO KOYANAGI,⁵ NAOKI YAMAMOTO,⁵ WANDA F. CANAS-FERREIRA,⁴
YAW ADU-SARKODIE,⁶ and TAKESHI KURATA¹

ABSTRACT

The phylogenetic variability of part of the long terminal repeat (LTR) region of HIV-2 strains isolated in 1995 from five individuals residing in Bissau, the capital city of Guinea-Bissau, and collected from seven persons from Kumasi, Ghana in 1996–1997, was analyzed. All Guinean samples and all but one Ghanaian sample clustered with HIV-2 subtype A. One Ghanaian sample (14%) was classified as HIV-2 subtype B. This study adds to previous reports on HIV-2 subtype distribution in West Africa indicating local prevalence of HIV-2 subtype B in Ivory Coast and neighboring Ghana.

HUMAN IMMUNODEFICIENCY VIRUS TYPE 2 (HIV-2) is common in West Africa (Ivory Coast, Ghana, Guinea-Bissau, Senegal, The Gambia, Cape Verde Islands, Mali)^{1,2} and in India.³ In addition, HIV-2 infection has been documented mainly in African immigrants.⁴ In geographic regions where both HIV-1 and HIV-2 are prevalent, their spread has been unequal. Studies have shown that HIV-2 is perinatally and sexually less transmissible than HIV-1.^{5,6} Although HIV-2 AIDS appears to be similar to HIV-1-induced AIDS, the rate of disease progression in HIV-2-infected individuals is much slower than in those with HIV-1.^{6,7} The ratio of HIV-1 versus HIV-2 infections is rapidly increasing over time. In Ghana, Hishida *et al.* reported that HIV seroprevalence of HIV/AIDS patient or suspected cases in 1990–1992 for HIV-1, HIV-2, and dual infection were 65, 21, and 14%, respectively.⁸ According to the data of 1999 HIV sentinel surveillance in Ghana, HIV-1, HIV-2, and dual infection rates were 92.8, 2.8, and 4.4%, respectively.⁹ Seven HIV-2 genetic subtypes, A to G, have been reported to date.^{10–12} HIV-2 subtype A viruses have been documented in diverse locations

across western Africa^{1,2} as well as in Europe,⁴ India,³ and South Korea.¹³ Subtype B exhibits a more restricted geographical distribution and has been reported mainly in Ivory Coast¹⁴ and Ghana,¹⁵ with a few cases documented in Europe¹⁶ and the Middle East.¹⁷ Subtypes C, D, E, F, and G are each represented by single sequences that were obtained from HIV-2-seropositive individuals living in rural areas of Liberia (subtypes C and D), Sierra Leone (subtypes E and F), or Ivory Coast (subtype G).^{10–12}

The present study focuses on HIV-2 subtype distribution among HIV-2-seropositive individuals in Guinea-Bissau ($n = 5$) and Ghana ($n = 7$). Ninety-four Guinean individuals attending an outpatient clinic at the Tropical Medicine Center of Bissau, the capital city of Guinea-Bissau, in 1995 were screened for HIV-1 and HIV-2 antibodies by enzyme-linked immunosorbent assay (ELISA) (Innotest HIV-1/HIV-2 Ab sp; Innogenetics, Zwijnaarde, Belgium). Reactive specimens were subsequently confirmed by Western blot analysis (New Lav Blot 1–2; Sanofi Diagnostics Pasteur, Marnes-la-Coquette,

¹National Institute of Infectious Diseases, AIDS Research Center, Tokyo 162-8640, Japan.

²Noguchi Memorial Institute for Medical Research, Ghana University, Accra, Ghana.

³Institute of Tropical Medicine, 2000 Antwerp, Belgium.

⁴Unidade de Virologia, Instituto de Higiene e Medicina Tropical (UNL), Lisbon P-1349-008, Portugal.

⁵Tokyo Medical and Dental University, Tokyo 113-8519, Japan.

⁶School of Medical Sciences, Kumasi, Ghana.

France). Twenty-one of 94 (22.3%) Guinean individuals tested positive for HIV-2. Eighteen of 21 Guinean samples were confirmed HIV-2 positive by nested polymerase chain reaction (PCR) using long terminal repeat (LTR) and/or *env* primers (primers L100/L200 and L101/L201 for LTR, and primers SE24/SE25bis and SE28/SE27bis for *env*) as described previously.¹⁸

Five Guinean HIV-2-positive samples and seven samples isolated from HIV-2-positive individuals from Ghana (Kumasi) in 1996–1997 were used for subtyping purposes. Genomic DNA was extracted from primary peripheral blood mononuclear cells (PBMCs) of HIV-2-seropositive individuals. A 324-base pair fragment encoding part of the LTR region (nucleotides 60 to 383 according to HIV-2 ROD; accession number X05291) was PCR amplified as described previously.¹⁸ PCR products were purified with a QIAquick gel extraction kit (Qiagen, Valencia, CA). The recovered PCR products were subjected to direct sequencing in both directions, using an ABI PRISM dye terminator cycle sequencing Ready Reaction kit (Applied Biosystems, Foster City, CA) and an automatic sequencer (ABI model 373A; Applied Biosystems). The newly determined HIV-2 LTR sequences were aligned with 15 previously documented sequences of HIV-2 isolates representing HIV-2 subtypes A, B, and G, for which LTR sequence information is currently avail-

able. Distance calculation, tree construction, and bootstrap analysis were realized with the software package TREECON, as previously described.¹⁹

Phylogenetically, all Guinean samples and six (86%) Ghanaian samples are classified as HIV-2 subtype A; one Ghanaian sample (14%; NJ205) belongs to HIV-2 subtype B. Within subtype A, four Ghanaian samples (NJ42, NJ79, NJ206, and NJ207) strongly clustered with HIV-2 ROD, previously isolated from a person of the Cape Verde Islands (Fig. 1). Our results indicate that at least two different subtypes of HIV-2, subtype A and subtype B, cocirculated in Kumasi, Ghana between 1990 and 1997. So far only HIV-2 subtype A has been found in Guinea-Bissau.²⁰ In Ghana, cocirculation of HIV-2 subtypes A and B has already given rise to HIV-2 A/B recombinants.¹⁵

This study adds to the previous reports on HIV-2 subtype distribution in Guinea-Bissau and Ghana. Regarding HIV-2 subtype distribution, so far subtype B has mainly been identified in Ivory Coast, where the majority of HIV-2 infections are due to subtype B (71%, 20 of 28),¹⁴ and to less extent in Ghana. The reason for this local prevalence of HIV-2 subtype B is unclear. There are currently no studies indicating differences in pathogenesis and transmissibility for HIV-2 subtypes. However, data on molecular epidemiology of HIV-2 subtypes remain scarce, and on the basis of our current knowledge it is

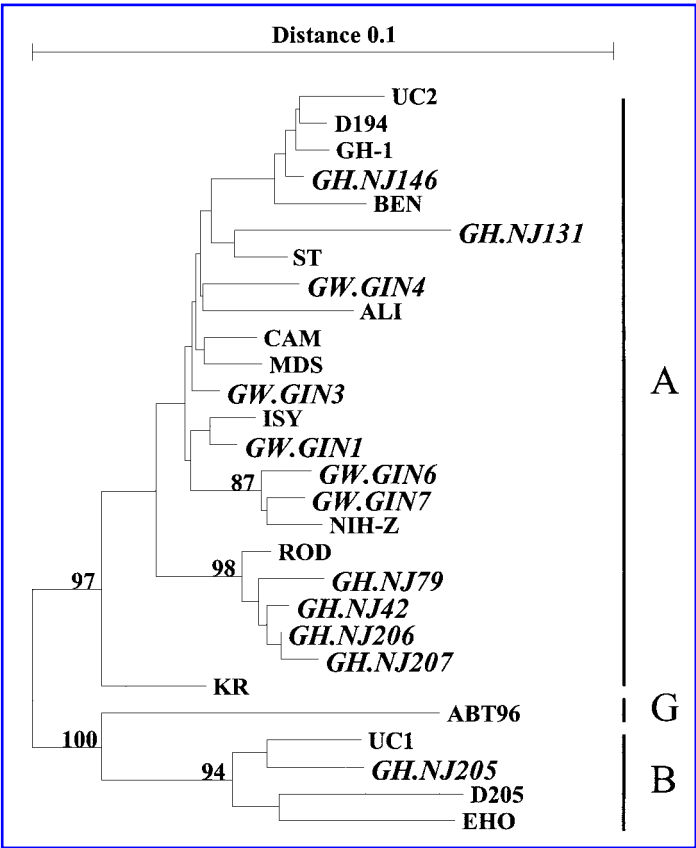


FIG. 1. Phylogenetic tree based on 346 unambiguously aligned positions of 28 HIV-2 sequences encoding part of the LTR. Sequences determined in this study are represented in italic. A total of 1000 bootstrap samples were analyzed. Bootstrap values are given in percentages at the internodes if they exceed the 70% level. The distance between two sequences is obtained by summing the lengths of the connecting branches by using the scale at the top. The tree is rooted arbitrarily. Strain names of samples from Guinea-Bissau and Ghana identified in this study are preceded by country codes GW and GH, respectively.

speculative to address evolutionary advantage to either subtype. As the earliest samples documented for HIV-2 subtype distribution in Ivory Coast were of both subtype A and B, sharing a similar range of diversity, it seems plausible to believe that the relative success of subtype A in most West African countries is due to a founder effect. Further studies of HIV-2 isolates may reveal the differences in biological properties between HIV-2 subtypes A and B.

ACKNOWLEDGMENTS

We acknowledge the staff of the Virology Unit, Noguchi Memorial Institute for Medical Research. This work was partially supported by the Japanese Human Science Foundation (Tokyo, Japan) and the Japan International Cooperation Agency (JICA).

SEQUENCE DATA

The HIV-2 LTR nucleotide sequence data were deposited in the EMBL, GenBank, and DDBJ nucleotide sequence databases under the following accession numbers: AY039114–AY039125.

REFERENCES

1. Miyazaki M: Epidemiological characteristics of human immunodeficiency virus type-2 infection in Africa. *Int J STD AIDS* 1995; 6:75–80.
2. Peeters M, Koumare B, Mulanga C, *et al.*: Genetic subtypes of HIV type 1 and HIV type 2 strains in commercial sex workers from Bamako, Mali. *AIDS Res Hum Retroviruses* 1998;14:51–58.
3. Grez M, Dietrich U, Balfe P, *et al.*: Genetic analysis of human immunodeficiency virus type 1 and 2 (HIV-1 and HIV-2) mixed infections in India reveals a recent spread of HIV-1 and HIV-2 from a single ancestor for each of these viruses. *J Virol* 1994;68:2161–2168.
4. Matheron S, Mendoza-Sassi G, Simon F, Olivares R, Coulaud JP, and Brun-Vezinet F: HIV-1 and HIV-2 AIDS in African patients living in Paris. *AIDS* 1997;11:934–936.
5. Kanki PJ, Travers KU, MBoup S, *et al.*: Slower heterosexual spread of HIV-2 than HIV-1. *Lancet* 1994;343:943–946.
6. Whittle H, Morris J, Todd J, *et al.*: HIV-2-infected patients survive longer than HIV-1-infected patients. *AIDS* 1994;8:1617–1620.
7. Pepin J, Morgan G, Dunn D, *et al.*: HIV-2-induced immunosuppression among asymptomatic West African prostitutes: Evidence that HIV-1 is pathogenic, but less so than HIV-1. *AIDS* 1991; 5:1165–1172.
8. Hishida O, Ayisi NK, Aidoo M, *et al.*: Serological survey of HIV-1, HIV-2 and human T-cell leukemia virus type 1 for suspected AIDS cases in Ghana. *AIDS* 1994;8:1257–1261.
9. Ministry of Health, Ghana: *AIDS Surveillance (January to December 1999)*. National AIDS/STD Control Programme, Ministry of Health—Ghana, Accra, Ghana.
10. Gao F, Yue L, Robertson DL, *et al.*: Genetic diversity of human immunodeficiency virus type 2: Evidence for distinct sequence subtypes with differences in virus biology. *J Virol* 1994;68:7433–7447.
11. Chen Z, Luckay A, Sodora DL, *et al.*: Human immunodeficiency virus type 2 (HIV-2) seroprevalence and characterization of a distinct HIV-2 genetic subtype from the natural range of simian immunodeficiency virus-infected sooty mangabeys. *J Virol* 1997; 71:3953–3960.
12. Yamaguchi J, Devare SG, and Brennan CA: Identification of a new HIV-2 subtype based on phylogenetic analysis of full-length genomic sequence. *AIDS Res Hum Retroviruses* 2000;16:925–930.
13. Kim SS, Kim EY, Park KY, *et al.*: Introduction of human immunodeficiency virus 2 infection into South Korea. *Acta Virol* 2000; 44:15–22.
14. Pieniazek D, Ellenberger D, Janini LM, *et al.*: Predominance of human immunodeficiency virus type 2 subtype B in Abidjan, Ivory Coast. *AIDS Res Hum Retroviruses* 1999;15:603–608.
15. Takehisa J, Osei-Kwasi M, Ayisi NK, *et al.*: Phylogenetic analysis of HIV type 2 in Ghana and intrasubtype recombination in HIV type 2. *AIDS Res Hum Retroviruses* 1997;13:621–623.
16. Heredia A, Vallejo A, Soriano V, *et al.*: Genetic analysis of an HIV type 2 subtype B virus from a Spanish individual with AIDS. *AIDS Res Hum Retroviruses* 1997;13:899–900.
17. Pieniazek D, Baggs J, Hu DJ, *et al.*: Introduction of HIV-2 and multiple HIV-1 subtypes to Lebanon. *Emerg Infect Dis* 1998;4: 649–656.
18. Ishikawa K, Fransen K, Ariyoshi K, *et al.*: Improved detection of HIV-2 proviral DNA in dually seroreactive individuals by PCR. *AIDS* 1998;12:1419–1425.
19. Van de Peer Y and De Wachter R: TREECON for Windows: A software package for the construction and drawing of evolutionary trees for the Microsoft Windows environment. *Comput Appl Biosci* 1994;10:569–570.
20. Esteves A, Parreira R, Piedade J, Venenno T, and Canas-Ferreira WF: Genetic characterization of HIV type 1 and type 2 from Bissau, Guinea-Bissau (West Africa). *Virus Res* 2000;68:51–61.

Address reprint requests to:

Koichi Ishikawa

AIDS Research Center

National Institute of Infectious Diseases

1-23-1 Toyama Shinjyuku, Tokyo, Japan

E-mail: kishikaw@nih.go.jp

This article has been cited by:

1. Jean Ruelle, Mahamoudou Sanou, Hsin-Fu Liu, Anne-Thérèse Vandenbroucke, Armelle Duquenne, Patrick Goubau. 2007. Genetic Polymorphisms and Resistance Mutations of HIV Type 2 in Antiretroviral-Naive Patients in Burkina Faso. *AIDS Research and Human Retroviruses* **23**:8, 955-964. [[Abstract](#)] [[Full Text PDF](#)] [[Full Text PDF with Links](#)]
2. Lucia Fischetti, Ohene Opare-Sem, Daniel Candotti, Francis Sarkodie, Helen Lee, Jean Pierre Allain. 2004. Molecular epidemiology of HIV in Ghana: Dominance of CRF02_AG. *Journal of Medical Virology* **73**:2, 158-166. [[CrossRef](#)]