Journal of Clinical Virology 81 (2016) 53–57

Contents lists available at ScienceDirect

Journal of Clinical Virology

journal homepage: www.elsevier.com/locate/jcv

The trend of transmitted drug resistance in newly diagnosed antiretroviral-naive HIV/AIDS patients during 1999–2012 in South Korea

Mina Park, Mee-Kyung Kee, JeeEun Rhee, Ji-Young Park, Sung Soon Kim, Chun Kang, Ju-yeon Choi*

Division of AIDS, Department of Immunology and Pathology, National Institute of Health South Korea, The Korea Centers for Disease Control and Prevention, Republic of Korea

ARTICLE INFO

Article history:
Received 22 March 2016
Received in revised form 27 May 2016
Accepted 1 June 2016

Keywords:
Transmitted resistance
HIV-1 *pol*
NRTI
NNRTI
PI

ABSTRACT

Background: The use of antiretroviral drugs has reduced the mortality and morbidity of patients with HIV/AIDS. More than 20 antiretroviral drugs have been used in patients with HIV/AIDS since zidovudine was first introduced in 1991 in South Korea.
Objectives: To investigate and estimate the annual prevalence of transmitted drug resistance and drug-resistant variants of HIV-1 in newly diagnosed antiretroviral-naive patients in South Korea during 1999–2012.
Study design: Plasma specimens were collected from 928 antiretroviral-naive patients during 1999–2012. Mutations in the protease and reverse transcriptase sections of the HIV-1 *pol* gene were identified using the Stanford HIV Drug Resistance Database (Stanford DB).
Results: Among 928 HIV-1 isolates from antiretroviral-naive patients, 45 (4.8%) showed 'intermediate' or 'resistant' drug resistance. The predicted prevalence of drug resistance among isolates was 2.2%, 2.7%, and 0.3% for resistance to nucleoside reverse transcriptase inhibitors, non-nucleoside reverse transcriptase inhibitors, and protease inhibitors, respectively.
Conclusions: There was no significant increase in the prevalence of drug resistance among antiretroviral-naive patients infected with HIV-1 during 1999–2012 in South Korea, although there was a slight increase during 2009–2012. The emergence of drug-resistant variants will continue to be monitored by national surveys.

1. Background

More than 30 U.S. Food and Drug Administration-approved antiretroviral drugs have been used for the treatment of human immunodeficiency virus and acquired immune deficiency syndrome (HIV/AIDS) patients worldwide [1,2]. The mortality and morbidity of patients with HIV/AIDS has been dramatically decreased by highly active antiretroviral therapy (HAART), but problems related to antiretroviral drug resistance still occur [3,4]. The appearance of drug-resistant HIV-1 variants withmutations of antiretroviral drug target genes is the main obstacle to the effectiveness of HAART.

In South Korea, the cumulative number of newlydiagnosed HIV/AIDS patients was reported to be 9410 during 1985–2012 by the Korean Centers for Disease Control and Prevention (KCDC). The public health centers have operated HIV/AIDS patients' management and care program since HIV was first introduced to Korea in the mid-1980s. Approximately 20 antiretroviral drugs have been used and about 60 hospitals treat HIV/AIDS patients in South Korea [5]. Antiretroviral therapy was introduced as zidovudine alone or a combination of 2 nucleoside reverse transcriptase inhibitors (NRTIs) in the late 1990s [6–8]. HAART protocols including protease inhibitors (PIs) have been used in South Korea since 1997 [9]. Since 2011, new antiretroviral drugs such as the non-nucleoside reverse transcriptase inhibitor (NNRTI) etravirine and the PI darunavir have

* Corresponding author at: Division of AIDS, Department of Immunology and Pathology, National Institute of Health South Korea, The Korea Centers for Disease Control and Prevention, 187 Osongsaengmyeong2-ro, Osong-eup, Heungduk-gu, Cheongju-si, Chungcheongbuk-do 28159, Republic of Korea.
E-mail address: juyeon.choi.hb@gmail.com (J.-y. Choi).

http://dx.doi.org/10.1016/j.jcv.2016.06.001
1386-6532/

Table 1
Demographic characteristics of the Korean antiretroviral-naive HIV-1-infected population during 1999–2012 (n = 928).

Antiretroviral-naive patients[c] (n = 928)									
Characteristics	Year								
	1999–2005 (n = 300)[d]	2006–2012 (n = 628)	2006 (n = 39)	2007 (n = 70)	2008 (n = 119)	2009 (n = 70)	2010 (n = 64)	2011 (n = 125)	2012 (n = 141)
No. of annually reported cases[a]	2593	5583	749	740	797	768	773	888	868
Gender, total n (%)[b]	300 (11.6)	628 (11.4)	39 (5.2)	70 (9.5)	119 (14.9)	70 (9.1)	64 (8.3)	125 (14.1)	141 (16.2)
Male	279	586	35	67	110	67	58	116	133
Female	21	42	4	3	9	3	6	9	8
Age (years)									
Mean	38.3	41.1	45.5	40.5	39.8	40.1	41.1	42.4	39.3
Range	15–71	1–80	22–77	17–65	17–68	19–70	2–72	18–75	1–80
Subtype									
B	288	576	37	67	108	67	61	111	125
non-B	12	52	2	3	11	3	3	14	16
Transmission risk category									
Heterosexual contact	159	252	12	42	51	26	26	44	51
Homosexual contact	118	200	18	22	46	24	24	30	36
Vertical transmission	–	2	–	–	–	–	1	–	1
Unidentified	23	174	9	6	22	20	13	51	53
Plasma HIV RNA level									
Mean log copies/mL	5.42	4.92	5.00	4.87	4.90	4.96	4.85	5.02	4.87
No. of tested patients	175	628	39	70	119	70	64	125	141
Rangeof log copies/mL	2.45–7.26	3.04–>7.00	3.15–6.32	3.04–6.48	3.09–7.00	3.23–>7.00	3.26–6.40	3.18–>7.00	3.21–6.58

[a] Number of annual cases means the total number of cases reported to KCDC by hospitals in South Korea.
[b] Values in parentheses show the number of tested cases divided by the number of reported cases. Variations in the HIV-1 *pol* gene have been monitored continuously by using a subset of more than 10% of the samples obtained from newly diagnosed antiretroviral-naive patients every year since 1999 in South Korea.
[c] These samples were taken from patients within 1 year after the confirmation of their HIV-1 diagnosis.
[d] Reference (5).

been incorporated into HAART in South Korea. And, genotypic drug resistance assays requested by more than 60 Korean hospitals were performed for free by the Korea National Institute of Health (KNIH) during July 2004–December 2014, and the prevalence of drug resistance in antiretroviral-naive patients has been monitored since 1999.

The average prevalence of drug resistance for newly diagnosed patients infected with HIV-1 was very low (4.3%) during 1999–2005 [5]. Since then, there have been several studies on antiretroviral drug resistance and mutation sites of HIV-1 in South Korea [10–13].

2. Objectives

In this study, we aimed to determine the annual prevalence of transmitted drug resistance in newly diagnosed antiretroviral-naive patients in South Korea during 1999–2012. To achieve this, we sequenced the protease (PR) and reverse transcriptase (RT) sections of the HIV-1 *pol* gene in plasma samples from 928 antiretroviral-naive patients in South Korea.

3. Study design

3.1. Study population and HIV-1 RNA extraction

The Plasma samples derived from 928 antiretroviral-naive patients were collected for genotypic drug resistance testing at KNIH during 1999–2012 from newly diagnosed patients with HIV-1 infection who had not received antiretroviral therapy. Variations in the HIV-1 *pol* gene were monitored continuously using a subset of about 10% of these samples each year (Table 1). HIV-1 RNA was extracted using an automated sample preparation system (m2000sp; Abbott Molecular, Des Plaines, IL, USA) and used for genotypic assays. Data were anonymized before analysis and all indications were converted into designated labels at the KCDC. The institutional review board "KCDC Research Ethics Committee (no. 2012-05CON-11-P)" approved this study.

3.2. Polymerase chain reaction (PCR) amplification and sequencing

The PCR and reverse transcription-PCR (RT-PCR) conditions were based on the laboratory protocol from the Center for AIDS Research at Stanford University for sequencingthe PR and RT sections of the HIV-1 *pol* gene. RT–PCR and a nested PCR reaction were performed as described [5]. The PCR product of *pol* (about 1.3 KB) containing the entire PR and RT sequences was subjected to direct sequencing using ABI PRISM®BigDye™ Terminator Cycle Sequencing Ready Reaction Kits (Applied Biosystems, Foster City, CA, USA) in an automated sequencer (ABI PRISM®3110, Applied Biosystems) after purification using gel electrophoresis and PCR Cleanup Kits (Millipore, Madison, WI, USA).

3.3. Analysis of drug resistance

The *pol* nucleotides and encoded amino acid sequences were aligned using the EditSeq and MegAlign programs in the Lasergene software package (version 5.06; DNASTAR, Madison, WI, USA). Interpretation of drug resistant mutations was identified by the consensus statement from the Stanford database (DB) for HIV PR (codons 1–99) and RT (codons 1–300). In addition, the Stanford HIVdb program was used to analyze the subtype and calculate the level of resistance against each antiretroviral drug (release notes for HIVseq, HIVdb, and HIValg are available at http://hivdb.stanford.edu/). The intensity of drug resistance was classified as S (susceptible, potential low level resistance); I (low or intermediate level resistance) or R (high level resistance), based on the Stanford DB HIValg results.