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Four-class drug-resistant HIV-1 subtype C in a treatment experienced individual on dolutegravir-based antiretroviral therapy in Botswana

Kaelo K. Seatla^{a,b,f}, Ava Avalos^{d,f,h}, Sikhulile Moyo^{a,e}, Madisa Mine^{a,d}, Thabo Diphoko^{a,b}, Mosepele Mosepele^{a,c,f}, Tendani Gaolatlhe^{a,c}, Christopher F. Rowley^e, Dinah Ramaabya^d, Joseph N. Jarvis^{a,c,f,g}, Ishmael Kasvosve^b, Simani Gaseitsiwe^{a,e}

^aBotswana Harvard AIDS Institute Partnership;

^bDepartment of Medical Laboratory Sciences, Faculty of Health Sciences;

^cDepartment of Medicine, Faculty of Medicine, University of Botswana;

^dMinistry of Health and Wellness, Gaborone, Botswana;

^eDepartment of Immunology & Infectious Diseases, Harvard T.H. Chan School of Public Health, Boston, Massachusetts, USA;

^fInfectious Disease Care Clinic, Princess Marina Hospital, Gaborone, Botswana;

^gDepartment of Clinical Research, Faculty of Infectious and Tropical Diseases, London School of Hygiene and Tropical Medicine, London, UK;

^hCareena Centre for Health, Gaborone, Botswana.

Abstract

There are limited data on the effectiveness of dolutegravir (DTG)-based combination antiretroviral therapy (ART) in real-life settings in southern Africa where HIV-1 subtype C predominates. We report a patient infected with HIV-1 subtype C on DTG-based ART previously exposed to raltegravir who developed multidrug resistance mutations to four antiretroviral classes. There is need for drug resistance monitoring and clinical vigilance to ensure effectiveness of HIV treatment programs even in the era of DTG-based ART.

Maintaining viral load suppression in people living with HIV (PLWHIV) is critical to ensure both the health of PLWHIV and prevent onward transmission of HIV to sexual partners [1–3]. Despite major advances in the development of antiretroviral (ARV) drugs and ARV therapy (ART) treatment guidelines [4–6], low and middle-income countries (LMICs)

Correspondence to Dr Kaelo K. Seatla, MBBS, DTMH, Botswana Harvard AIDS Institute Partnership, Private Bag BO320, Bontleng, Gaborone, Botswana. Tel: +267 390 2671; fax: +267 390 1284; kseatla@bhp.org.bw, kaeloseatlamd@yahoo.com.

Author information: K.K.S. is a PhD student at the University of Botswana funded by SANTHE.

Authors' contributions: K.K.S. wrote the first article, is involved in the clinical care of the patient and performed most of the drug resistance experiments. A.A., M.M., T.G., J.N.J., K.K.S. were and are still involved in the clinical care of the patient. T.D., S.M., D.R., C.F.R., M.M., J.N.J., I.K., S.G. performed the drug resistance testing and/or provided supervision and guidance. All authors helped to edit and approved the final article version for publication.

Conflicts of interest

There are no conflicts of interest.

continue to face challenges such as poor ART adherence, limited HIV care specialists, drug stock-outs, lack of ancillary healthcare services and so on [7–9]. These barriers to effective ART may lead to development of extensive HIV-1 drug resistance.

Dolutegravir (DTG) has recently been introduced as part of the first-line ART regimen in Botswana and may soon be adopted by other countries in sub-Saharan Africa (SSA) [10,11]. Therefore, there is a need to monitor the development of integrase strand transfer inhibitor (INSTI) resistance mutations. We report a case of a four-class drug-resistant HIV-1 subtype C in a 51-year-old man currently on DTG-based ART with persistent viremia.

Chart reviews of treatment-experienced patients not virologically suppressed while on salvage ART therapy were conducted at a local tertiary hospital as part of the Botswana Epidemiological ART Treatment (BEAT) cohort study. A case-file reported herein was identified and an analysis of the patient's medical record was conducted with information gathered from September 2003 to November 2017. The patient provided written informed consent to complete the analysis for publication, and the BEAT study is approved by the human research and development council of Botswana.

Genotypic resistance testing (GRT) was performed using Sanger sequencing of the *pol* and *env* genes. Sequences obtained were assessed for drug resistance mutations using the Stanford University HIV Drug Resistance Database (<https://hivdb.stanford.edu>), International Antiviral Society USA 2017 mutational list [12] and coreceptor usage was determined using with geno2pheno_[coreceptor] [13]. All sequences generated were submitted to GenBank under accession numbers MG989439-MG989443, MH004049 and MH004050.

The patient was initiated on zidovudine (AZT), lamivudine and nevirapine on September 2003 as per the 2002 Botswana National ART guidelines. Apart from persistent viremias, his 14-year follow-up history has been clinically uneventful and devoid of opportunistic infections.

He intermittently achieved virological suppression between September 2003 and April 2014 (six HIV-1 RNA levels <400 copies/ml out of 67 tests) (Fig. 1). His ART regimens while virologically suppressed were AZT/tenofovir (TDF)/ritonavir-boosted lopinavir from April 2007 and TDF/emtricitabine/ritonavir-boosted darunavir/raltegravir for the remaining aviremic episodes from February 2010. The majority of viral load revealed virological failure with viral load ranging between 2.61 log₁₀ and 5.88 log₁₀ copies/ml (Fig. 1). Nine switches in ARV medications were made from September 2003 to November 2017 (Fig. 1). Adherence to clinic appointments and medications was suboptimal, and he experienced one episode of drug stock-out of ARVs (darunavir) for approximately 2 weeks in March 2012. His social circumstances remained challenging, making it difficult for him to be adherent to his medications despite multiple counseling and adherence support sessions from healthcare providers.

The patient is infected with HIV-1 subtype C clade and, during the period under review, had a median HIV-1 viral load of 3.48 log₁₀ (Q1, Q3: 3.27, 3.68) copies/ml and absolute CD4⁺ T-cell count of 595 cells/μl (Q1, Q3: 501, 692), respectively. As of November 2017, viral load and CD4⁺ T-cell count were 3.57 log₁₀ copies/ml and 670 cells/μl, respectively (Fig. 1).

GRT analysis revealed major drug resistance mutations conferring resistance to nucleoside (nucleotide) reverse transcriptase inhibitors (K65R, D67N, K70R, M184V and K219N), nonnucleotide reverse-transcriptase inhibitors (Y181C), protease inhibitors (V32I, I47V, I54L, I84V) and INSTIs (E138K, G140A, S147G, Q148R, T97A). The virus was chemokine receptor 5 tropic and did not have any resistance mutations to enfuvirtide (T-20).

To our knowledge, this is the first report in SSA of a patient with a virus which has developed drug resistance mutations to all the standard ARVs belonging to four classes including INSTIs. Multidrug-resistant mutations likely developed as a result of multiple factors, including suboptimal adherence, inadequate psychosocial support and limited HIV specialist care - all issues encountered frequently in LMICs.

Although in-vitro and in-vivo clinical studies have shown DTG to retain some activity against a virus with INSTI mutations [14,15], this is not always the case in real-life settings as highlighted by this case report. An interesting observation made was the isolated emergence of the INSTI accessory resistance mutation T97A between May 2015 and November 2017 and concurrent rise in viral load from 2888 to 3690 copies/ml between the same time period. Similar observations of viral rebound after sole emergence of T97A mutations have recently been reported [16]. Phenotypic testing, ARV drug tracing levels and directly observed therapy would have been ideal but often these are not available or feasible in LMICs.

The patient may benefit from a regimen that includes two new agents expected to be fully active, such as maraviroc (MVC) or enfuvirtide (T-20) with an optimized background regimen. Currently, MVC and T-20 are not available under the Botswana National HIV Clinical Care Guidelines.

As this patient remains sexually active with significant psychosocial issues, this raises important public health issues including the possible introduction of a multidrug-resistant HIV-1 subtype C strain into the circulating pool.

The case highlights the need for continued monitoring of HIV-1 viral load and the development of robust HIV drug resistance surveillance systems for failing and highly treatment-experienced patients. Managing treatment-experienced patients in LMICs, in which there is often inadequate psychosocial support, lack of clinical HIV care expertise, limited ARV options and drug resistance testing capacity, will remain an ongoing challenge that will need to become a public health priority to ensure the effectiveness of national HIV programs.

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Fig. 1. Plasma viral loads, CD4⁺ T-cell count and HAART regimens at different time points over a 14-year period for the patient.

Horizontal line within chart depicts viral load of 400 copies/ml. Numbered rectangular callouts depict when drug resistance testing was done. (1) Mar 2009; [†]nucleotide reverse-transcriptase inhibitors, D67N, K70R, M184V; nonnucleotide reverse-transcriptase inhibitors, Y181C; protease inhibitors major resistance mutations, V32I, I47V, I54L, I84V, [†]protease inhibitors accessory resistance mutations, L33F, G73V, L89T; integrase strand transfer inhibitors, not tested. (2) Apr 2009; [†]nucleotide reverse-transcriptase inhibitors, D67N, K70R, M184V, K219N; [†]nonnucleotide reverse-transcriptase inhibitors, Y181C; [†]protease inhibitors major resistance mutations, V32I, I47V, I54L, I84V, protease inhibitors accessory resistance mutations, L33F, G73V, L89T; integrase strand transfer inhibitors, not

tested. (3) May 2015; [†]nucleotide reverse-transcriptase inhibitors, K65KR, D67DN, K70KR, M184MV, K219KHNQ; [†]nonnucleotide reverse-transcriptase inhibitors, none; [†]protease inhibitors major resistance mutations, V32VI, I54IL, I84IV, [†]protease inhibitors accessory resistance mutations, L33LF, G73GV, L89IMT; [†]integrase strand transfer inhibitors major resistance mutations, E138K, G140A, Q148R, integrase strand transfer inhibitors accessory resistance mutations, none; (4) Aug 2016; [†]nucleotide reverse-transcriptase inhibitors, D67N, K70R, M184V; [†]nonnucleotide reverse-transcriptase inhibitors, none; [†]protease inhibitors major resistance mutations, V32I, I47V, I54L, I84V, [†]protease inhibitors accessory resistance mutations, L33F, G73V, L89T; integrase strand transfer inhibitors, not tested. (5) Nov 2017; [†]integrase strand transfer inhibitors major resistance mutations, E138K, G140A, S147G, Q148R, integrase strand transfer inhibitors accessory resistance mutations, T97A; [‡]fusion inhibitors, none; entry inhibitors, [§]R5 tropic; nucleotide reverse-transcriptase inhibitors, not tested; nonnucleotide reverse-transcriptase inhibitors, not tested. 3TC, lamivudine; ABC, abacavir; AZT, zidovudine; d4T, didanosine; ddI, didanosine; DRV/r, ritonavir boosted darunavir; DTG, dolutegravir; EI, entry inhibitor; FI, fusion inhibitor; FTC, emtricitabine; INSTI, integrase strand transfer inhibitor; LPV/r, ritonavir boosted lopinavir; NFV, nelfinavir; NNRTI, nonnucleotide reverse-transcriptase inhibitor; NRTI, nucleoside/nucleotide reverse transcriptase inhibitor; NVP, nevirapine; PI, protease inhibitor; RAL, raltegravir; RM, resistance mutation; SQV/r, ritonavir-boosted saquinavir; TDF, tenofovir; VF, virological failure. Drug resistance mutations interpreted using [†]Stanford University HIV Drug resistance database; [‡]International Antiviral Society USA 2017 mutational list; [§]coreceptor usage assessed with geno2pheno_[coreceptor]. *Source*: modified version from <http://bioafrica.mrc.ac.za/workshops/PDFs/deOliveiraRegaDB.pdf>.