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Increasing HIV-1 Pre-Treatment Drug Resistance among Antiretroviral-Naïve Adults Initiating Treatment between 2006 and 2014 in Nairobi, Kenya

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Summary

Antiretroviral-naïve adults initiating antiretroviral therapy (**ART**) in Nairobi, Kenya were tested for HIV-1 drug resistance at codons K103N, Y181C, G190A, M184V, and K65R using an oligonucleotide ligation assay (**OLA**). Prevalence of pre-treatment drug resistance (**PDR**) increased from 3.89% in 2006 to 10.93% in 2014 ($p<0.001$), and 95% of those with resistance had at least one non-nucleoside reverse transcriptase inhibitor (**NNRTI**) mutation. Resistance to tenofovir (K65R) was found in 2014 but not in 2006.

Keywords

transmitted drug resistance; oligonucleotide ligation assay; HIV; Kenya; antiretroviral therapy

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This data was presented at the 2014 International Workshop on Antiviral Drug Resistance held June 3–7, 2014 in Berlin, Germany (Oral Abstract #1).

Since the widespread introduction of antiretroviral (**ARV**) therapy (**ART**) in sub-Saharan Africa there has been concern that HIV-1 drug resistance will become prevalent. Cross-sectional studies have found varying levels of pre-treatment HIV drug resistance (**PDR**) among ARV-naïve adults that is associated with earlier ART rollout on the continent [1]. This study enrolled ARV-naïve adults at two separate time points in 2006 and 2014 at a single clinic site in Nairobi, Kenya and measured PDR using an oligonucleotide ligation assay (**OLA**).

Data and specimens were collected from ARV-naïve participants enrolled in two clinical trials in 2006 and 2014 at the Coptic Hope Center for Infectious Diseases in Nairobi [2, 3]. Sociodemographic information, medical records, and blood samples were analyzed, including baseline pre-treatment plasma (2006) and peripheral blood mononuclear cells (**PBMC**) (2014). Following quantification of amplifiable copies of HIV cDNA and DNA by realtime PCR [4], ≥100 HIV templates from each participant underwent nested PCR of HIV *pol* to generate amplicons for OLA. OLA examined point mutations at K103N, Y181C, G190A, and M184V across all specimens [5]. K65R was evaluated by OLA in 2014 and pyrosequencing was performed on enrollment plasma from those with subsequent virologic treatment failure in 2006 [4]. The proportion of mutant in each subject's HIV population was quantified by comparing optical densities to standards containing 0%, 2%, 5%, 10%, 25%, 50%, 75% or 100% mutant, with PDR defined by ≥2% mutant [4].

Wilcoxon rank sum and Chi-square tests were used to compare continuous and categorical characteristics. The prevalence of PDR was compared between 2006 and 2014 using Poisson regression with robust variance. Stata SE v11 (Statacorp, College Station, TX, USA). The study was approved by institutional review boards at the University of Washington (Seattle, WA, USA), Seattle Children's Hospital (Seattle, WA, USA), and Kenyatta National Hospital (Nairobi, Kenya).

A total of 953 adults were examined: 386 in 2006 and 567 in 2014. Marital status, number of years educated, and age of sexual debut were similar between the two cohorts, and the proportion of women in 2006 and 2014 was 66% and 60% respectively. The median age of those in 2014 was slightly older than in 2006 (38 vs. 36 years; $p<0.01$), with higher economic status (10% vs. 33% unemployment; $p<0.01$), better housing (60% vs. 47% flush toilet access; $p<0.01$), fewer lifetime sexual partners (3 vs. 4 partners; $p<0.01$), and higher median CD4 cell count at baseline (198 vs. 115 cells/mL; $p<0.01$).

The prevalence of PDR was 3.89% [95% Confidence Interval (CI), 2.19%–6.33%] in 2006 and 10.93% (95% CI, 8.49%–13.80%) in 2014. The unadjusted prevalence of PDR in 2014 was 2.81 times greater than in 2006 (95% CI, 1.62, 4.87, $p<0.001$). Adjusting for age, gender, marital/attached status, unemployment, lifetime number of sexual partners, travel time to clinic, and CD4 count (n=917), the prevalence ratio of PDR was 3.11 (95% CI: 1.69, 5.73, $p<0.001$).

In 2006, 24 PDR mutations were detected in 15 participants at baseline (Table 1) [4]. Fifteen of the participants had mutations only to NNRTI (K103N, Y181C, G190A), and three had mutations to NNRTI plus lamivudine (M184V). The median mutant frequency within a

subject's viral population was 82% (range, 2%–100%). In 2014, 84 mutations were detected in 62 participants. Fifty-eight had mutations to NNRTI, two had only lamivudine mutations, and two had only tenofovir (K65R) mutations. The median mutant frequency within a subject's viral population was 32% (range 2%–100%).

All ARV-naïve adults with PDR in 2006 and 94% in 2014, had at least one NNRTI mutation (overall, 95%). Eighty-seven percent of cases with a lamivudine mutation had a concomitant NNRTI and/or tenofovir mutation across both cohorts. Resistance to tenofovir was detected in 2014 when it was found in 10% (6 of 62) of those with PDR and 1.1% (6 of 576) of those initiating ART (Table 1).

The prevalence of PDR among ARV-naïve adults nearly tripled, rising from 3.9% to 10.9%, between 2006 and 2014 in Nairobi, Kenya, and 95% of those with resistance had at least one NNRTI mutation. While not found in 2006, PDR to tenofovir was present in 1.1% of all individuals initiating ART in 2014, two years after tenofovir replaced stavudine as part of first-line ART regimens in Kenya.

This study examined HIV drug resistance among a large sample of ARV-naïve adults across nearly a decade. Providing ART at no cost, the clinic attracts a broad patient population from diverse sociodemographic backgrounds representative of the capital [6]. The serial nature of these data from this large clinic adds a meaningful dimension to previous cross-sectional studies [7, 8], and describes significant resistance trends over time, rather than generalizing results across East Africa [9].

There are several weaknesses in this study. We did not measure PDR during acute/early HIV infection but at ART initiation, and while NNRTI-resistance does not rapidly revert back to wild type [10], this study may underestimate resistance, especially the 2006 genotypes from plasma. The study only surveyed drug resistant mutations at codons K103N, Y181C, G190A, M184V, and K65R, which has the potential to underestimate, not overestimate the prevalence of PDR. Testing PDR in PBMC by OLA and defining resistance at ≥2% detects mutant frequencies below those discernable by consensus sequencing [11]; however, low frequency mutations are associated with greater virologic failure and therefore remain clinically relevant [4]. Despite recruitment at one clinic, several sociodemographic differences existed between the cohorts and likely reflected Kenya's economic development. While only ARV-naïve adults were examined in this study, previous exposure to ART was based on self-report and could be subject to misclassification. Finally, although OLA for K65 was not performed among all 386 participants in 2006, pyrosequencing performed on the 54 participants with subsequent virologic failure found no K65R in this cohort [4].

In summary, we found that ARV-naïve adults initiating ART in 2014 were three times more likely to have PDR than those initiating ART in 2006. This increased prevalence of PDR suggests that approximately one out of ten ARV-naïve adults in Kenya could fail their first-line ART [4].

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M Chung designed and implemented the study, supervised the on-site data management, interpreted the data, and wrote the paper. R Silverman performed the statistical analysis, interpreted the data, and helped write the paper. I Beck conducted the laboratory analysis, interpreted the data, and helped write the paper. N Yatich helped implement the study and interpret the data. S Dross conducted the laboratory analysis, interpreted the data, and helped write the paper. J McKernan-Mullin helped conduct the laboratory analysis and interpret the data. S Bii helped conduct the laboratory analysis and interpret the data. K Tapia analyzed the data and helped interpret the results. J Stern analyzed the data and helped interpret the results. B. Chohan helped implement the study and manage the laboratory. S Sakr helped implement and design the study. J Kiarie helped implement the study, oversee data collection, and helped write the paper. L Frenkel led the laboratory analysis, designed the study, interpreted the data, and wrote the paper.

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References

1. Aghokeng AF, Kouanfack C, Laurent C, et al. Scale-up of antiretroviral treatment in sub-Saharan Africa is accompanied by increasing HIV-1 drug resistance mutations in drug-naive patients. AIDS. 2011; 25:2183–8. [PubMed: 21860346]
2. Chung MH, Richardson BA, Tapia K, et al. A randomized controlled trial comparing the effects of counseling and alarm device on HAART adherence and virologic outcomes. PLoS Med. 2011; 8:e1000422. [PubMed: 21390262]
3. Chung MH, Drake AL, Richardson BA, et al. Impact of prior HAART use on clinical outcomes in a large Kenyan HIV treatment program. Curr HIV Res. 2009; 7:441–6. [PubMed: 19601781]
4. Chung MH, Beck IA, Dross S, et al. Oligonucleotide ligation assay detects HIV drug resistance associated with virologic failure among antiretroviral-naive adults in Kenya. J Acquir Immune Defic Syndr. 2014; 67:246–53. [PubMed: 25140907]
5. Micek MA, Blanco AJ, Beck IA, et al. Nevirapine resistance by timing of HIV type 1 infection in infants treated with single-dose nevirapine. Clin Infect Dis. 2010; 50:1405–14. [PubMed: 20384494]
6. Chung MH, Kohler P, Attwa M, Thiga J, John-Stewart GC. Comparing clinic retention between residents and nonresidents of Kibera, Kenya. J Acquir Immune Defic Syndr. 2010; 53:422–4. [PubMed: 20190592]
7. Hamers RL, Wallis CL, Kityo C, et al. HIV-1 drug resistance in antiretroviral-naive individuals in sub-Saharan Africa after rollout of antiretroviral therapy: a multicentre observational study. Lancet Infect Dis. 2011; 11:750–9. [PubMed: 21802367]
8. Hassan AS, Mwaringa SM, Obonyo CA, et al. Low Prevalence of Transmitted HIV Type 1 Drug Resistance Among Antiretroviral-Naive Adults in a Rural HIV Clinic in Kenya. AIDS Res Hum Retroviruses. 2013; 29:129–35. [PubMed: 22900472]
9. Gupta RK, Jordan MR, Sultan BJ, et al. Global trends in antiretroviral resistance in treatment-naive individuals with HIV after rollout of antiretroviral treatment in resource-limited settings: a global collaborative study and meta-regression analysis. Lancet. 2012; 380:1250–8. [PubMed: 22828485]
10. Little SJ, Frost SD, Wong JK, et al. Persistence of transmitted drug resistance among subjects with primary human immunodeficiency virus infection. J Virol. 2008; 82:5510–8. [PubMed: 18353964]
11. Ellis GM, Mahalanabis M, Beck IA, et al. Comparison of oligonucleotide ligation assay and consensus sequencing for detection of drug-resistant mutants of human immunodeficiency virus type 1 in peripheral blood mononuclear cells and plasma. J Clin Microbiol. 2004; 42:3670–4. [PubMed: 15297515]

Table 1

Prevalence of pre-antiretroviral treatment HIV drug resistance mutations detected by OLA in each antiretroviral-naïve adult by cohort year.*

Mutant Codon	2006 (N=386)	2014 (N=567)	Total (N=953)
K103N	8 (2.1%)	30 (5.3%)	**38 (4.0%)**
Y181C	1 (0.3%)	8 (1.4%)	**10 (0.9%)**
G190A	0 (0%)	3 (0.5%)	**3 (0.3%)**
M184V	0 (0%)	2 (0.4%)	**2 (0.2%)**
K65	0 (0%)	2 (0.4%)	**2 (0.2%)**
K103N + Y181C	0 (0%)	1 (0.2%)	**1 (0.1%)**
K103N + G190A	1 (0.3%)	5 (0.9%)	**6 (0.6%)**
K103N + M184V	1 (0.3%)	3 (0.5%)	**4 (0.4%)**
Y181C + G190A	1 (0.3%)	0 (0%)	**1 (0.1%)**
Y181C + M184V	0 (0%)	1 (0.2%)	**1 (0.1%)**
Y181C + K65R	0 (0%)	1 (0.2%)	**1 (0.1%)**
G190A + M184V	0 (0%)	1 (0.2%)	**1 (0.1%)**
K103N + Y181C + M184V	2 (0.5%)	2 (0.4%)	**4 (0.4%)**
K103N + G190A + Y181C	1 (0.3%)	0 (0%)	**1 (0.1%)**
Y181C + M184V +K65R	0 (0%)	2 (0.4%)	**2 (0.2%)**
G190A + M184V + K65R	0 (0%)	1 (0.2%)	**1 (0.1%)**
Total	**15 *(3.9%)**	**62 *(10.9%)**	**77 (8.1%)**

*Poisson regression with robust standard error estimates comparing 2014 to 2006 found an unadjusted prevalence ratio of 2.81 (95% CI, 1.62, 4.87; p<0.001).