A14 Estimating time since HIV infection using next-generation sequencing data: A unique tool to help understand HIV prevention among high-risk young women in Ukraine

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The transitions study examines HIV risk among adolescent girls and young women through their sexual life course from first sex, to past and current engagement in casual sex, transactional sex, and, for some, formal sex work (FSW). Understanding the timing of HIV infection and the circumstances around early infection in young females is critical to HIV prevention interventions. We inferred time since HIV infection using next-generation sequencing (NGS) of the HIV *pol* gene isolated from cross-sectional samples among high-risk young women in Dnipro, Ukraine. Dried blood spots were collected on Whatman 903 cards from young women aged 14–24 engaged in casual sex ($n = 894$), transactional sex ($n = 464$), and FSW ($n = 452$). The HIV *pol* gene was sequenced using an in-house NGS HIV drug resistance mutation genotyping assay. Time since HIV infection was inferred using an online tool as described by Puller et al. (2017) freely available at https://hiv.biozentrum.unibas.ch/ETI/. The mean estimated time since HIV infection (ETI) for participants engaged in casual sex, transactional sex, and FSW is 1.98, 1.84, and 3.01 years, respectively. ETI was used to determine the duration of HIV infection for each participant and compared to the number of sexually active years prior to FSW. Among FSW, 61 per cent of participants were infected with HIV prior to entry into sex work. In general, ETI from NGS data suggests that FSWs were infected with HIV before entry into FSW. Expansion of targeted prevention programs beyond FSW could play an important role in mitigating HIV transmission at the population level.

A15 Archived ART resistance in the latent reservoir of virally suppressed Ugandans

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The increased access of antiretroviral therapy (ART) has drastically improved the health of infected individuals. However, increased levels of ART resistance globally threaten ART effectiveness. Resistance monitoring is currently limited to viremic individuals or prior to ART initiation. The archival nature of the HIV latent reservoir (LR) of virally suppressed patients allows examination for the persistence of ART-resistant latent viral variants. Whole blood samples were collected longitudinally in Rakai, Uganda, from 70 virally suppressed HIV-1 infected individuals. The quantitative viral outgrowth assay was performed to measure the frequency of replication-competent latently infected resting-memory CD4 + (rCD4) T-cells. RNA was extracted from HIV p24-positive outgrowth supernatant, and the reverse transcriptase (RT) region was sequenced using a validated site-specific next-generation sequencing assay (Illumina, San Diego, CA). Consensus sequences containing >2.5 per cent of the total raw amplicons of each outgrowth well were analyzed for ART drug resistance mutations using the Stanford Database. The presence of clonal sequence is expressed as both percent clonality and Shannon Entropy. Replication-competent virus was cultured from 52/70 (74.3%) individuals, of which, RT-pol sequence data were obtained from 49/52 (94.3%) individuals. The presence of ART-resistant virus was found in the LR from one individual on second-line therapy that included a protease inhibitor. There were 20 and 44 total prominent consensus sequences from all wells at years 1 and 3 of follow-up, respectively. ART-resistant mutations for both RT-inhibitor drug classes were found in 30 per cent and 27.3 per cent of the total prominent consensus sequences of this one individual from years 1 and 3, respectively. The major ART resistance profile in this individual included: M184V, Y188L, K191E, and G190A. The percentage of total outgrowth that was clonal (percent clonality) increased from year 1 to year 3 (38.1–81.8%) and Shannon Entropy decreased (0.722–0.576). The presence of archived replication-competent ART-resistant virus in the LR was found in only one individual. There were two ART-resistant prominent consensus sequences isolated at year 3 that were not sampled 2 years earlier. The persistence of resistant, intact replication-competent proviral sequences in the LR of this individual seem to be supported by clonal expansion.

A16 Next-generation sequencing to detect transmitted drug resistance mutations in Romanian people who inject drugs

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Romania has faced an HIV outbreak among people who inject drugs (PWID) since 2011. The introduction of so-called 'legal highs' (amphetamine-type stimulants) on the drug market a few years prior contributed substantially to this outbreak. Next-generation sequencing (NGS) provides the possibility to detect drug resistance mutations with higher sensitivity than Sanger sequencing. The aim of this study was to search for transmitted drug resistance (TDR) mutations in strains from PWID recently diagnosed with HIV infection by parallel use of Sanger sequencing and NGS. The study was conducted on strains from 34 PWID diagnosed with HIV infection between 2016 and 2017. Sequencing was performed for the *pol* (PR, RT, and INT) and *env* (V2-V3 loop) regions. Sanger sequencing was performed with the commercial ViroseqTMHIV-1 Genotyping system (Abbott Laboratories) and with an in-house protocol for the *env* gene. NGS was performed in the same genomic regions using Nextera DNA Library Preparation Kit (Illumina) and the Miseq instrument (Illumina). NGS data were processed for error correction, read mapping, and detection of drug resistance mutations with HIV-1 Deepchek analysis software. Geno2pheno algorithm was used for viral tropism prediction and the WHO 2009 list for TDRM analysis. By using NGS, we detected seven cases (20.6%) of TDR in PWID and only two cases (5.8%) with standard sequencing. The TDR mutations detected by NGS were K103N, K101EN, Y181C, T215S in RT gene, I54V and M46L in PR, and none in INT. Two NNRTI mutations (K103N and K101EN) were detected in the same sample. Most of the TDR identified were present in the minority population (between 1% and 2% of the total reads) explaining the higher sensitivity of NGS method compared with standard sequencing. No significant differences were observed between these two methods when tropism prediction was analyzed. The majority of the viruses circulating in this group were R5-tropic. All strains showed more resistance mutations when analyzed by deep sequencing than by Sanger sequencing and more than previously observed in other risk groups. NGS proved to be a sensitive tool to detect TDR in newly infected PWID.

A17 The effect of intra-host evolution of HIV-2 capsid on disease progression

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The human immunodeficiency virus type 2 (HIV-2) is an important cause of acquired immune deficiency syndrome (AIDS) in West Africa. The virus started circulating in humans around 1938 and has spread predominantly within West Africa with an estimated 1–2 million people being infected today. Compared with the