Abstract

The advent of antiretroviral treatment (ART) has resulted in a dramatic reduction in AIDS-related morbidity and mortality. However, the emergence and spread of antiretroviral drug resistance (DR) threaten to negatively impact treatment regimens and compromise efforts to control the epidemic. It is recommended that surveillance of drug resistance occur in conjunction with scale-up efforts to ensure that appropriate first-line therapy is offered relative to the resistance that exists. However, standard resistance testing methods used in Sub-Saharan Africa rely on techniques that do not include low abundance DR variants (LADRVs) that have been documented to contribute to treatment failure. The use of next generation sequencing (NGS) has been shown to be more sensitive to LADRVS. We have carried out a preliminary investigation using NGS to determine the prevalence of LDRVS among a drug-naive population in North Rift Kenya. Antiretroviral-naive patients attending a care clinic in North Rift Kenya were requested to provide and with consent provided blood samples for DR analysis. DNA was extracted and amplified and nested PCR was conducted on the pol RT region using primers tagged with multiplex identifiers (MID). Resulting PCR amplicons were purified, quantified, and pyrosequenced using a GS FLX Titanium PicoTiterPlate (Roche). Valid pyrosequencing reads were aligned with HXB-2 and the frequency and distribution of nucleotide and amino acid changes were determined using an in-house Perl script. DR mutations were identified using the IAS-USA HIV DR mutation database. Sixty samples were successfully sequenced of which 26 were subtype A, 9 were subtype D, 2 were subtype C, and the remaining were recombinants. Forty-six (76.6%) had at least one drug resistance mutation, with 25 (41.6%) indicated as major and the remaining 21 (35%) indicated as minor. The most prevalent mutation was NRTI position K219Q/R (11/46, 24%) followed by NRTI M184V (5/46, 11%) and NNRTI K103N (4/46, 9%). Our use of NGS technology revealed a high prevalence of LADRVs among drug-naive populations in Kenya, a region with predominantly non-B subtypes. The impact of these mutations on the clinical outcome of ART can be ascertained only through long-term follow-up.