(77.55%) strains; and in 96.3% of subgenotype C (26/27) isolates. Stop codon mutation on Pre-Core, G1896A, was found in 23 of 98 patients (23.47%); among them, 20 of 23 (86.96%) are genotype B. Mutations related to antiviral drug resistance with frequency more than 1% in the reverse transcriptase (RT) domain of Polymerase gene were identified. 83.67% sequences have more than one mutation. 50% sequences have mutation at rtI169V/M, and all belong to sub-genotype B. Mutations at rtM204I/V and rtM250I were present in 23.47% and 11.22% of isolates (23 and 11 of 98, respectively), and was higher in sub-genotype C (P-values = 0.06 and <0.01, respectively). Our study provides high precision whole genome sequences of the Vietnamese HBV population and shows potential information related to treatment prediction for HBV infection as well as viral evolution. A further study with larger sample size and intensive analysis combined with clinical information promises more useful data for treatment and invention.

A18 Random amplification with next-generation sequencing to cover HIV and HCV full-length genomes

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Since both HIV and HCV are known to be highly variable and infections are treated with combination therapies that target different viral proteins, random amplification followed by next-generation sequencing could be an attractive alternative to Sanger sequencing for obtaining complete viral genomes. Moreover, one experimental approach could enable the simultaneous mapping of virus and human origin sequence reads, which is of particular interest for the detection of single nucleotide polymorphisms (SNPs) associated with disease progression and treatment response. A plasma sample obtained from a patient infected with HIV-1 recombinant subtype B and F1, and diagnosed with AIDS; and another sample from an HCV infected patient who progressed to fibrosis stage F1 were included in this pilot study. They were both characterized by a high viral load, i.e. >10M copies/ml for HIV and >3.5M IU/ml for HCV. Samples were directly extracted or pre-treated with homogenisation, centrifugation and filtration. Subsequently, purified nucleic acids were subjected to targeted (HIV-1 PR-RT and HCV NS3 protease) amplification and Sanger sequencing, or to random amplification followed by Illumina sequencing (~2M paired end reads attributed to each sample). Quality of the Illumina reads was checked using FastQC, followed by a *de novo* assembly of sample-specific contigs using the VICUNA and V-FAT software packages. In total, 0.3% ($n = 5557$) and 0.02% ($n = 421$) of the reads mapped to the HCV genome, respectively with or without virus enrichment steps. This sufficed for the *de novo* generated contigs to cover 98.3 and 51.3% of the HCV genome with and without pre-treatment steps, respectively. Lower numbers of reads could be mapped for HIV-1 (821 versus 196 without virus enrichment), resulting in only 45.6 and 21.7% coverage of the full-genome. For both samples, the number of reads (<2M) was too low to map SNPs of interest within the human genome. Subject to adaptations in the protocol, such as improved enrichment strategies and deeper sequencing, this approach could be suitable for the surveillance of local HIV-1 and HCV epidemics. More extensive studies are required to investigate its usefulness in clinical research.

A19 Genetic markers for protease inhibitor drug resistance in regions outside of the protease gene

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There are currently 6.8 million individuals infected with HIV-1 in South Africa. Nearly half of these individuals are on antiretroviral therapy. Protease inhibitors (PIs) form part of the South African national treatment guidelines as a first-line regimen for paediatric patients and a second-line regimen for adults. However, successful treatment is often hindered by the development of drug resistance. Resistance to PIs is characterised by a stepwise accumulation of mutations in the protease gene. However, studies have shown a low frequency of mutations in patients failing PIs. Regions outside of the protease, such as gag and more recently env, have been associated with PI drug resistance in the absence of protease mutations. We aim to identify genetic markers in the gag and envelope genes which are associated with PI treatment failure. Stored plasma samples from HIV-1-infected patients failing PI-based therapy ($n = 500$) will be collected from collaborators in South Africa and whole genome sequences from PI-naïve patients will be downloaded from the Los Alamos Sequence Database. Failure will be defined as having two consecutive viral loads >1,000 cpm after being on a PI-based regimen for ≥6 months. The whole HIV-1 genome will be amplified from the PI failures and sequenced using Illumina Miseq. Sequences will be aligned to a reference and analysed for single nucleotide polymorphisms (SNPs) in all HIV genes using Geneious v 8.1.8 and GATK software application. Genome wide association analysis using PLINK will be performed to identify SNPs in PI treated patients that are associated with treatment failure. Episodic directional selection model such as MEDS and IFEL will be used to identify mutations that occur at specific amino acid positions and confer resistance to PIs. The previously mentioned methods allow the identification of drug resistance mutations without the need of baseline samples. Preliminary data from GWAS analysis on 26 patients failing PI-based therapy has identified codons in envelope and gag which are significantly associated with PI failure. Further examination of these sites in the viral minority population has shown that they are present at >2% of the viral population. By increasing our sample size we will obtain a more comprehensive and robust analysis of the role of regions outside the protease gene in PI resistance. Analysis of the control samples is currently being performed.