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Background
To detect HIV minority populations of resistant virus, a hyper sensitive test is needed, and today’s Gold Standard, Sanger sequencing, is not able to detect minority population less than 10 to 20 percent of the virus population. Ultra-deep sequencing on the Miseq instrument allows the identification of HIV nucleotide variants and variant haplotype signatures present at <1% in samples. In this study, samples were sequenced on the Miseq and uploaded onto BaseSpace to be analyzed through the DeepChek HIV Pipeline. (Fig. 1).

Results
► Compatible with all the key genomic regions for drug resistance testing of HIV (Reverse transcriptase, Protease, Integrase, GP41 and GP120/V3) in either BAM of FASTQ formats, several types of analyses were performed like:
  ▪ a high resolution subtyping of each individual read
  ▪ a population variants calling
  ▪ a fully automated reporting.
► The outcomes of those data processing steps were then interpreted for drug resistance determination using a panel of guidelines part of the system.
► Results were produced and formatted in a simplified manner, directly compatible with an optimal, reliable, accurate, cost effective interpretation of minor variants even below 1% though efficient workflow, for research applications.

Methods
► A new pipeline (Fig. 2) combining MiSeq-HIV (RUO) via BaseSpace as an alternative to Sanger sequencing, opens a new domain of analysing minor HIV variant detection to < 1% of the viruses’ population.
► HIV-1 samples were sequenced via a MiSeq instrument (allowing a sensitive and reliable identification of HIV nucleotide variants even below 1%). In this study, nucleotide sequences were automatically uploaded onto BaseSpace via MiSeq Reporter and analysed by the DeepChek-HIV (RUO) software application.
► Compatible with your own primer design for analysis of following genes, Reverse Transcriptase, Protease and Integrase.

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Conclusion
Tracking of viral population is very important as it can determine expressed dual infections in early stages of viral evolution, co-infection, or superinfection. This study gives insight on how to get started with an automated HIV pipeline combining ultra-deep sequencing on the MiSeq an end-to-end solution starting with an easy sample preparation, an efficient and reliable data analysis and interpretation and a comprehensive reporting of subtypes, mutations and HIV drug resistance prediction.