

# Precise Identification and Genome Recovery of Viral Pathogens Through a Non-Specific Target Virome Clinical Test

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## Abstract

Massive parallel sequencing techniques radically changed the diagnostic workflow by providing a quick and powerful tool for clinical diagnosis and precision medicine. The basis of rare diseases as well as clinically relevant mutations underlying some types of cancers have been precisely diagnosed and characterized using NGS techniques. In a similar way, etiological causes of infectious diseases, such as encephalitis, arboviroses and hepatitis have been precisely identified. In this work, we report the validation process of a new metagenomic protocol for unbiased identification of viral pathogens in clinical samples. The test is based on a two-step cDNA random amplification followed by Illumina sequencing. Data generated is used as input to a custom bioinformatic pipeline especially developed for delivering fast, accurate and clinician-friendly diagnosis. The validation process was performed according to College of American Pathologists (CAP) guidelines, evaluating accuracy, intra and inter-assay reproducibility, and limit of detection. Preliminary results show accuracy rates of 100% compared with standard PCR tests, total agreement among different assays and 104 virions/mL as limit of detection (similar to PCR limits). Moreover, we were able to recover high quality data to characterize almost complete or whole genomes from different viral pathogens with mean depth of coverage ranging from 100x to 8000x. Recovered genomes were also used to identify viral genotype and drug resistance variants, as well as to perform phylogenetics analysis. Altogether, these results show a multi-functional test to potentially replace several gold-standard molecular diagnosis techniques such as qualitative PCR and Sanger sequencing method in a unique protocol. We emphasize that the virome technique developed in this work is not limited to previous knowledge of the pathogen, allowing early detection of outbreaks due to novel pathogens and playing a key role in public health surveillance.

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