

DeepChek® Assays: Your best ally for HIV genotyping & Drug Resistance Determination

From Capillary Electrophoresis to Deep Sequencing: An Improved HIV-1 Drug Resistance Assessment Solution Using In Vitro Diagnostic (IVD) Assays and Software

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Objective:

The objective of this study was to evaluate the performances of CE-IVD assays for HIV-1 drug-resistance assessment both for target-specific and whole-genome sequencing, using standardized end-to-end solution platforms.

Method:

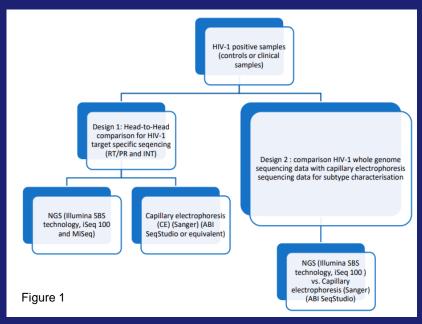


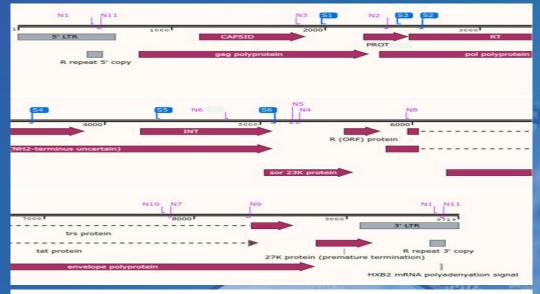
Figure 2

Localization of the CE and NGS primers for HIV drug resistance:

- In blue, CE primers for reverseprotease. transcriptase, integrase regions.
- In purple, NGS primers for the whole-genome HIV (Snapgene Software Version 5.25.5)

Table 1

A comparison of the NGS protocol and CE sequencing is summarized in Table 1. To process 24 samples (RT, PR, and INT) by CE sequencing, the time required for waiting, sample preparation, and overall time to result were 81.0 h, 2.0 h, and 83.0 h, respectively. The full workflow of database processing, analysis, and reporting using DeepChek®-HIV was more than 20 min per sample. To process 24 samples(RT, PR, and INT) by NGS, the time required for waiting, sample preparation, and overall time to result were 27.0 h, 4 h, and 31.0 h, respectively. The full workflow of database processing, analysis and reporting using DeepChek®-HIV was less than 2 min per sample.



Steps	NGS	Time/24 Samples (h)	CE	Time/24 Samples (h)
Sample preparation	RNA extraction kit	1.0	RNA extraction kit	1.0
Amplification	RT-PCR	4	RT-PCR	4
Purification Quantitation	Beads Purification	0.75	Enzymatic purification	0.2
	Quality control (TapeStation)	0.2	_	-
	Normalization (Qubit)	0.5	_	_
Library/se- quencing reac- tion	Library preparation	4	Sequencing reaction	2.5
Dilution Sequencing	Dilution and pooling Sequencing	20	Sequencing with SeqStudio 4-capillary	72
Data analysis	FastQ files DeepChek® using ANRS, HIVdb, etc.	0.2	ABI files DeepChek® using ANRS, HIVdb, etc.	1.0
Result	Handling time	4	Handling time	2
	Waiting time	27	Waiting time	81
	Time to result	31	Time to result	83
Price	Reagent cost \$/sample	100-150 *	Reagent cost/sample	80
Sensitivity		1 to 3%	- 1	20%

^{*} Including extraction, PCR, library preparation, indexes, sequencing, and software.

Conclusions:

The use of whole-genome sequencing is an additional and complementary tool to detect mutations in newly infected untreated patients and heavily experienced patients, both with higher HIV-1 viral-load profiles, to offer new insight and treatment strategies, especially using the new HIV-1 capsid/maturation inhibitors and to assess the potential clinical impact of mutations in the HIV-1 genome outside of the usual HIV-1 targets (RT/PR and INT).





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