Diagnostic accuracy of a near point-of-care HIV drug resistance test: a validation study of OLA-Simple in Kenya

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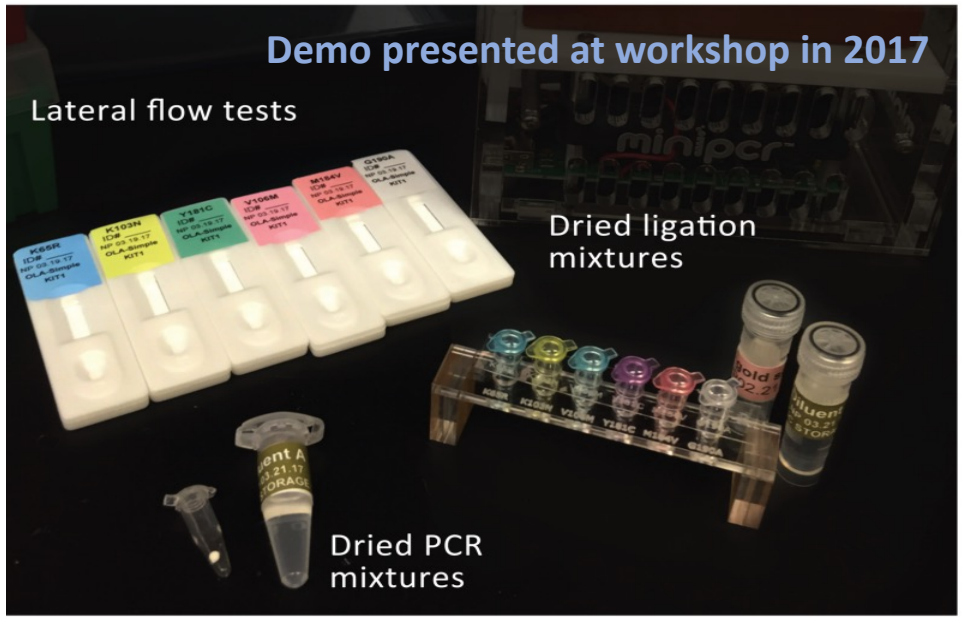
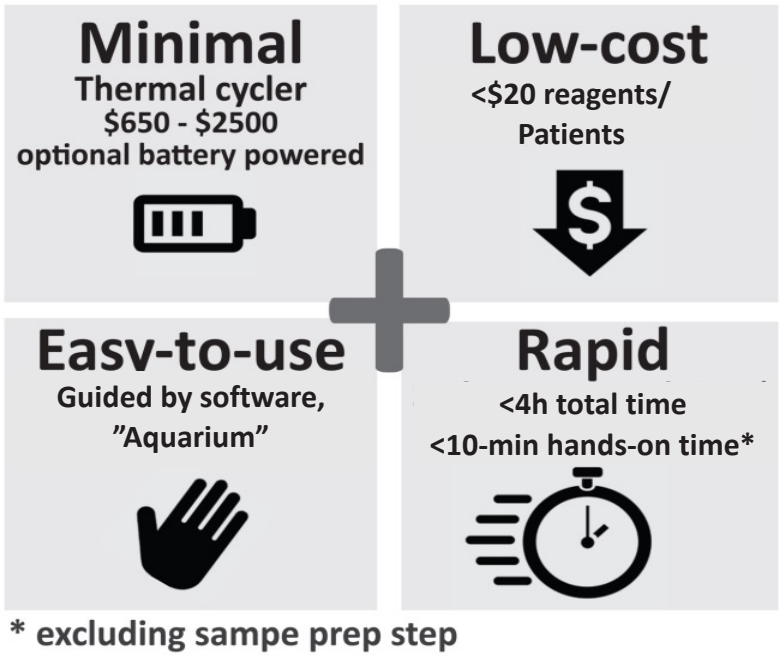
Background

OLA-Simple is **a low-cost, rapid, near point-of-care assay** platform that detects HIVDR mutations. It contains:

- ***Ready-to-go dried mixtures*** – easy assay set up
- ***Lateral flow tests*** – visual readout
- ***Interactive software "Aquarium"*** – 1st-time users previously showed 97% accuracy operating OLA-Simple [1,2]





This presented work: OLA-Simple probes were developed for **HIV-1 RT mutations** in **HIV subtypes A, B, C, D and AE**, Targeted mutation sites were selected based on HIVDR results from Sanger consensus sequencing in **children whose medications included abacavir.**

[1] N Panpradist, et.al. 2019. Ebiomedicine; [2] N Panpradist and J Vrana et al. 2021 PLOS Global Health

Methods

STUDY SITES

Study time frame: April-June 2022

PHASE I (3 days on-site training)

Total of seven lab technicians performed OLA-Simple on two blinded control specimens (negative plasma spiked with DNA).

PHASE II 8-10 weeks of independent testing by local lab technicians

Blind testing at KEMRI/CDC:
87 plasmas collected from children and pregnant/postpartum women

Blind testing at NHRL:
59 archived plasmas

OLA-Simple results for K65R, L74V/I, K103N/S, Y115F, Y181C, M184V, G190A blindly **classified by software** as mutant, wildtype, or indeterminate (excluded two samples with incorrect image result formats)

Compared to reference assays: Sanger sequencing and laboratory OLA. Laboratory OLA detects ≥2% low-frequency variants.

Results

- **Phase I results.** Genotype classifications of the results obtained both during the observed and unobserved runs had a **100% agreements** with the genotypes of the DNA controls.
- **Phase II results.**

Table 1. Summary of OLA-Simple results compared to Sanger with discordant results adjudicated by sensitive benchmark (laboratory OLA)

	K65R	L74V/I	K103N/S	Y115F	Y181C	M184V	G190A	Total
True (-)	114	96	50	112	103	35	88	**589**
True (+)	6	25	69	11	21	87	33	**258**
False (+)	2	3	8	0	2	2	6	**23**
False (-)	0	0	4	4	1	4	1	**14**
Indeterminate	10	8	1	5	5	4	4	**37**

- Of 146 plasma samples tested, OLA-Simple **successfully amplified** 134 specimens (**91.8%**).
- From successfully amplified specimens, 132 had OLA-Simple image data for analysis of 924 codons which included 275 mutant, 612 wild-type and **4% indeterminate results**.
- OLA-Simple detected **6** low-frequency mutant variants **missed by Sanger** (confirmed by laboratory OLA sensitive to 2% mutant); it **misclassified 2.6% wild-type codons as mutant** and **1.6% mutant codons as wildtype.**

Sensitivity
94.7% [95%CI: 91.3-97.1]

Specificity
96.3% [95%CI: 94.5-97.6%]

Indeterminate
4% [95%CI:2.8-5.5]

Summary

Conclusions:

- This in-field validation study serves as a significant step towards implementation of OLA-Simple in LMICs.
- It reveals OLA-Simple's 94.7% sensitivity and 96.3% specificity.

Next steps:

- Validation of prospective specimens
- Expanding OLA-Simple to detect dolutegravir mutations (Abstracts **23** and **60**).

Researchers at NHRL (I received permission to share their excitements via this photo)