Towards Rapid Dapivirine Measurement with a Portable, Low-Cost Platform

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Clinical trials have established a relationship between PrEP drug levels and effectiveness.
Need for Drug-Level Feedback (DLF)

Both Clinicians and PrEP Recipients have Expressed Desire for DLF
- Facilitate candid discussions
- Spur timely interventions

REACH Study
- DLF provided monthly with additional support options
- Higher adherence to the ring 73%–89%

Significant logistical hurdles to providing prompt DLF

56 days
Turnaround time

DLF available at 55% visits

Nair et al., 2023, Lancet HIV DOI:10.1016/S2352-3018(23)00227-8
Drug-Level Feedback Tools

Liquid Chromatography-tandem Mass Spectrometry
LC-MS/MS

- Expensive (>\$200/test)
- Long turn around time (>1 month)
- Requires highly trained personnel
- Bulky equipment in centralized labs

Drug-Level Feedback Tools
Urine Lateral Flow Assay

- Measures only recent adherence
- May be vulnerable to “white coat” effect
- Primarily focuses on tenofovir (TFV)-based regimens

Gandhi et al., 2019, J Acquir Immune Defic Syndr. DOI 10.1097/QAI.0000000000001971
Drain et al., 2020, JMIR Research Protocols DOI: 10.1016/j.eclinm.2018.09.003
Koenig et al., 2017, HIV Medicine DOI:10.1111/hiv.12518
REverSe TRanscrIptase Chain Termination Assay

Assay inspired by the mechanism of action of reverse transcriptase inhibitors

Dilute blood in water
For easy sample prep

Add components needed for DNA synthesis

Incubate at 37°C for 30 min

Add intercalating dye and fluorescence readout

TFV-DP = Tenofovir diphosphate

Olanrewaju et al., 2020 ACS Sensors DOI:10.1021/acssensors.9b02198
Olanrewaju et al., 2021 Virology Journal DOI:10.1186/s12985-021-01543-x
Olanrewaju et al., 2022 Bioengineering and Translational Medicine DOI:10.1002/btm2.10369
RESTRICT Measurements Correlate with LC-MS/MS Results

- Rapid test (30 min)
- Simple workflow
- High correlation with LC-MS/MS measurements

TFV-DP in Dried Blood Spots from people on oral PrEP
Samples are diluted and heated for easy sample prep

Craig et al., CROI 2022
Innovations in this Presentation

Expand RESTRICT to measure reverse transcriptase inhibitors used in extended-release HIV prevention and treatment

Validate use of portable reader suitable for point-of-care settings
RESTRICT Measures Clinically Relevant Concentrations of Dapivirine

Reported clinically relevant DPV concentrations by sample type

CVF=cervicovaginal fluid   Ring=residual ring levels

European Medicines Agency, Product Information
RESTRICT Measures Clinically Relevant Concentrations of Long-Acting Rilpivirine

Alert Threshold for Long-Acting Injectable Rilpivirine: 32ng/mL

French National Agency for Research on AIDS and Viral Hepatitis Emerging Infectious Diseases (ANRS)
Expanding RESTRICT to a Portable Platform

Harmony Reader

- Developed for point-of-care COVID-19 diagnostics
- Integrated portable heater and fluorescence reader

Harmony portable reader

Panpradist et al., 2021, Science Advances DOI:10.1126/sciadv.abj1281
Practical Advantages of Portable Reader

Needs Less Space
1% of the bench space

Easily Moved
40 kg vs 0.1 kg

Less Expensive
~$30,000 vs ~$300

Harmony portable reader compared to traditional benchtop plate reader (SpectraMax iD3)

Panpradist et al., 2021, Science Advances DOI: 10.1126/sciadv.abj1281
RESTRICT Workflow in Portable Reader

DNA synthesis reagents and drug sample

Fluorescent Dye

37 °C
30 min

Drug Concentration

Fluorescence (%)

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The Harmony and Plate Reader Output have Excellent Agreement in Buffer

![Graph showing fluorescence intensity vs. Dapivirine concentration for Plate Reader and Portable Reader.]

- **Fluorescence Intensity (%)**
- **Dapivirine Concentration (M)**

**Plate Reader** vs. **Portable Reader**

- **r=0.9807**
- **p<0.0001**
Summary and Future Work

RESTRICT can measure clinically relevant DPV concentrations in buffer on an inexpensive, portable reader.

Future work includes validation with clinical samples.
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