Development of a Point of Care Urine Tenofovir Test – A PrEP and ART Adherence tool

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Background

- There are 1.2 million people at high-risk of HIV infection in the US and millions worldwide.
- Tenofovir (TFV) is the major component of Pre-Exposure Prophylaxis (PrEP) for HIV prevention and a critical component of antiretroviral therapy (ART) for the treatment of HIV.
- For both the prevention and treatment of HIV, adherence is critical.
- Current adherence to TFV-containing regimens is sub-optimal, but effective methods for drug monitoring can improve adherence.
- There is a need for an affordable, disposable, easy to use point of care (POC) test to asses adherence in order to better manage PrEP and ART care.

Methods

Synthesized a novel TFV derivative to be conjugated to KLH and be used an immunogen to produce anti – TFV monoclonal antibodies

Screened and selected antibodies in a competitive enzyme linked immunoassay (ELISA) format to assess sensitivity and specificity

LFIA prototype was developed on a nitrocellulose pad with a sample port, antibody gold conjugate strip, immobilized TFV-BSA strip and a control

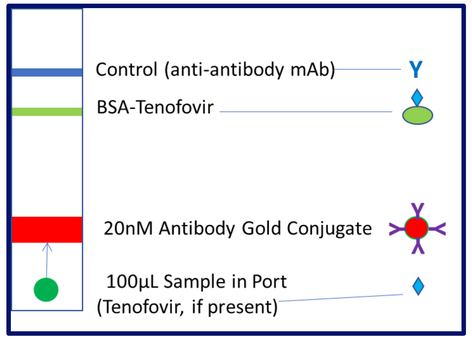
Varying spiked TFV concentration samples were added to the prototype and sensitivity was assessed

Objective

To establish a qualitative lateral flow immunoassay (LFIA) to measure the concentration of TFV in a patient's urine to measure and promote adherence.

Lateral Flow Immunoassay

Strip Components



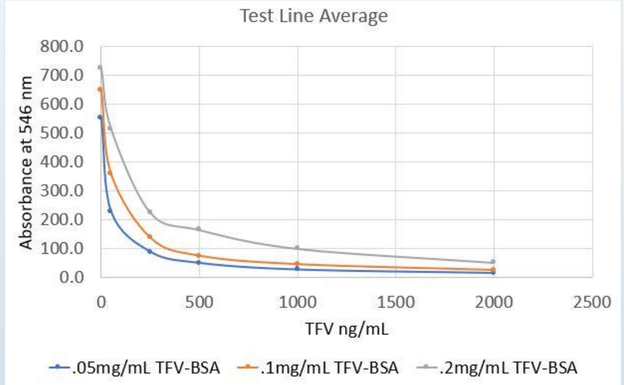
- The sample migrates upward starting from the sample port and ending at the control line.
- First, the sample is added to the port and then migrates to the antibody gold conjugate.
- If TFV is present in the sample, it will bind to the conjugate and not be captured at the immobilized TFV-BSA conjugate test line = no 1st line.
- If TFV is not present in the sample, the antibody conjugate will not bind and it will be captured at the immobilized TFV-BSA conjugate test line = 1st line.
- The control line (C) is the built-in procedural control, which consists of an anti-mouse IgG = 2nd line.

Results

mAB Sensitivity and Specificity of ELISA

	LC-MS (+)	LC-MS (-)
Antibody (+)	50	8
Antibody (-)	0	140

LFIA Prototype Standard Curve



- The monoclonal antibody performance in the competitive ELISA format demonstrated 100% sensitivity and 94.6% specificity.
- When the TFV-BSA strip was dipped in the gold nanoparticle antibody conjugate the results showed sensitive detection of TFV spiked samples across a dilution series.
- The prototype has a limit of detection of 1,250 ng/mL.

Conclusion

A POC LFIA prototype that could detect TFV concentrations in the urine matrix was developed and could prove to be a useful tool in promoting optimal adherence for HIV prevention and treatment.

Future Steps

- Optimization, validation, verification, and manufacturing initiation of the POC LFIA test.
- Clinical trials to assess the acceptability, feasibility, and effectiveness of the test in real world settings.
- Scale-up of the POC test to patients and providers worldwide.

References

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