PCR-Free Ultrasensitive Hiv And Other Virus Quantitation Device

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SUMMARY

UCLA researchers in the Department of Electrical Engineering & Bioengineering and Department of Medicine have developed a novel integrated device that can perform label-free ultrasensitive measurements of viruses in fluids (i.e. HIV in blood), obviating PCR and bulky, costly infrastructure required for current generation clinical assays.

BACKGROUND

Measurement of viral load is crucial to monitor that antiviral therapy is effective and that viral replication that could result in drug resistance mutations is shut down. Currently, PCR is the only accepted method to monitor for plasma viremia. This requires real-time PCR, which in turn requires RNA extraction, enzymatic reverse transcription, thermocycling with a thermostable polymerase and primers and a fluorescent probe, and laser fluorescence detection. These steps require expensive and fragile machinery and technical expertise, which is not practical in remote areas. Various versions of enzyme-linked immunosorbent assays (i.e. ELISAs) are technically simpler and require little specialized equipment, but are limited by sensitivity for low levels of proteins. While PCR-based detection can measure 10-25 viruses/mL in plasma, the best commercial ELISA can only detect 100,000 viruses/mL in plasma. Therefore, there is a need for sensitive, yet affordable device for viral load measurement.

INNOVATION

Researchers at UCLA have developed a novel device that can replace current PCR-based clinical assays for label-free ultrasensitive measurements of viruses in fluids. In one embodiment, this device can perform functions including: i) physical isolation of HIV virions from whole blood and separation from endogenous free anti-HIV antibodies; ii) lysis of virions to release its proteins including the Gag p24 antigen; iii) ultrasensitive quantification of p24 at <100 viruses/mL plasma, a level comparable to commercially utilized PCR-based assays for quantitating plasma viremia. Moreover, this device only requires 10 uL of sample loaded on the biosensor, which means that the volume of whole blood from a fingerstick would be more than sufficient to achieve this level of detection on a point-of-care device. The label-free approach allows the signal on the target biomolecules directly transduced and read without any labeling, eliminating labeling reagents, their conjugations, and tedious washing procedures. As an alternative to labeling-based methods, this method minimizes requirements for infrastructure (e.g. light source, microscope, optical reader), cost, personnel, and turnaround time for many clinical lab assays.

APPLICATIONS

Virus (i.e. HIV) quantitation point-of-care device

ADVANTAGES

- Allows p24 quantification with an ultrasensitive lower limit of detection without labeling
- One device exhibited a preliminary lower limit of detection of p24 of <10 fg/mL in buffer or human plasma, corresponding to a level of <100 viruses/mL plasma
- Only requires a very small volume of whole blood obtained from a fingerstick
- The label-free approach allows the signal on the target biomolecules directly transduced and read without any labeling, eliminating labeling reagents, their conjugations, and tedious washing procedures

STATE OF DEVELOPMENT

The label-free T-nanowire field-effect transistor biosensors exhibited a preliminary lower limit of detection of p24 of <10 fg/mL in buffer or human plasma, which corresponds to a level of <100 viruses/mL.

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OTHER INFORMATION

KEYWORDS

Medical device, label-free, sensitivity, virus quantitation, HIV, virus, PCR-free, clinical assay, viral load, plasma viremia monitoring, biosensor

CATEGORIZED AS

- **Biotechnology**
  - Health
- **Medical**
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  - Disease: Infectious Diseases
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