Rapid and Cost-effective Detection of Tuberculosis (TB)

S. Generelli, D. Migliorelli, H. Chai-Gao, S. Paoletti, T. Broger •

High costs are often among the major limitations to a widespread use of diagnostics tools at scale, especially in resource-limited regions. A simple, rapid, sensitive, biomarker-based, point-of-care TB diagnostic that makes use of an easily accessible sample like urine is considered to be one of the most urgently needed test by expert stakeholder groups. In collaboration with FIND [1] we are working on addressing this unmet need through the development of an assay for the rapid and highly-sensitive detection of Lipoarabinomannan (LAM) in urine using a combination of bead-based analyte concentration technology and detection with Screen Printed Electrodes (SPEs).

Two weeks of cough is a widely used symptomatic indicator to identify individuals with presumed active pulmonary tuberculosis (TB) who require diagnostic testing. Since most individuals with suspected TB do not have TB, a triage test can help narrow down the population that needs the more costly confirmatory testing [1]. A triage test is a simple, low-cost test for use by community health workers to rule out TB and direct individuals who require further evaluation (i.e. triage test positive) via a confirmatory test. Triage testing could take place at the same level of care as confirmatory testing especially in settings with a large influx of patients (e.g. crowded outpatient clinics), but typically would be done at lower levels of care (e.g. microscopy centre, primary care clinics, etc.). If the test turns out to be specific enough, it could even be used to diagnose TB and immediate treatment initiation (without confirmatory testing).

Commercially available point of care tests, based on lateral flow immunoassays have a too high detection limit, estimated to 500 pg/ml, not adapted for sensitive detection in patients suspected of having TB.

In order to respond to this unmet need, an improved LAM detection assay was developed in our labs based on antibodies provided by FIND. The target detection limit stated by FIND of 50 pg/ml was achieved, with a potential for further improvement to an estimated detection limit of less than 10 pg/ml (background +3 standard deviations) (Figure 1).

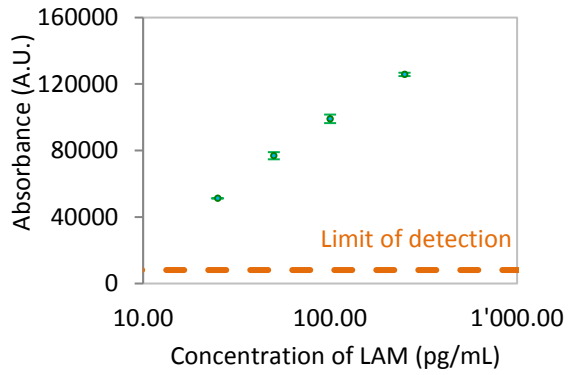

Figure 1: Calibration curve of LAM in PBS buffer.

The improved assay is being used as the basis for the development of a simple to use, cost-effective and rapid assay, compatible with the large-scale deployment in health care institutions.

CSEM has developed a process for diagnostics using Screen Printed Electrodes (SPEs) in combination with functionalized magnetic particles. This system was used in the past for the detection of TB biomarker ESAT-6 in a saliva matrix.

The process consists mainly of:

- Pre-concentration step: in order to collect the biomarkers of interest in a simple preparation procedure, functionalized magnetic particles were used. In addition the use of the chosen sample pre-concentration strategy allows the subsequent detection and quantification to be more sensitive and reach lower detection limits
- Detection step: Screen Printed Electrodes (SPEs) are cost effective, disposable devices that can be miniaturized and integrated in disposable microfluidic cartridges, if necessary, to work with sample volumes in the microliter range. SPEs will be used as the matrix to perform and quantify sandwich-type immunoassay.

The two pre-concentration and detection steps can be used in concomitance with each other, or used separately. For example, the pre-concentration step may be used as a pre-purification and pre-concentration step for the subsequent detection using commercially available ELISA detection kits or lateral flow immuno-chromatographic assays. This approach is applicable and adaptable for diagnostics on various body fluids as for example saliva, sputum, urine and of course blood.

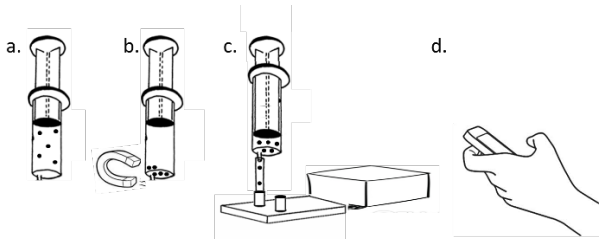

Figure 2: Schematic of the diagnostic assay. a) Sample collection and incubation; b) Pre-concentration; c) Injection in automated sensing device and further incubations; d) Data are sent to the user interface.

• FIND Switzerland

[1] www.finddx.org