Lipids to detect tuberculosis?

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Stop TB Partnership
New Diagnostics Working Group
Mycolic acids

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Different Mycobacteria have their own unique Mycolic Acid (MA) compositions
Mycolic acids

Three main classes of MA for *M. Tuberculosis*
Cord Factors (sugar esters)
(trehalose dimycolate, TDM)

We now have > 60 individual isomers to test!
Encouraging literature

Complex natural mixtures of cord factors can be used as an antigen for TB
(Yano et al/1991)

Isolated MA sub-classes are recognised by IgG antibodies
(Pan et al/1999)

Natural MA can be used as an antigen for TB in the presence of HIV-AIDS
(Verschoor et al/2002)

Can we improve on this with synthetic single isomers?
Different antigens give selective responses

Absorbance at 492 nm

Serum 1 TB+
Serum 2 and 3 TB-

Other microbial infections

Bovine - *M. Avium*

- Not infected
- Infected with TB Serum Sample
- Infected with M. Avium

Absorbance at 492 nm
ELISA results from a blind study of 350 WHO TB indigenous samples

Vaccination does not interfere

Different single synthetic MA and their derivatives give differential responses

Our best antigen gave 85% Sensitivity and 85% Specificity

UK negatives are much cleaner
Country variations!

By using two antigens with a set of Gambia samples we used a ‘traffic light’ system that gave

100 % Sensitivity

91 % Specificity
From a clinical setting to the field

We have taken 4 additional approaches

1. Evanesce
   On 350 blind samples
   73 % Sensitivity - 81 % Specificity

2. Impedance

Plus two PoC systems
Assay development

Au colloid

Coated with antigen

Antigen + Serum

Look for a change in the absorbance

Colour response within minutes

Positive  Negative
Sensor development

On 350 blind samples

89 % Sensitivity - 62 % Specificity
Lipid Antigens

Sera containing the antibodies of interest are added

Particle secondary antibody conjugate

Patent No. 1414369.7 – Filed 2014
Paper based device
# Paper based device

<table>
<thead>
<tr>
<th>TB positive serum</th>
<th>TB negative serum</th>
<th>Control (no serum)</th>
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<tbody>
<tr>
<td><img src="image1.jpg" alt="Image" /></td>
<td><img src="image2.jpg" alt="Image" /></td>
<td><img src="image3.jpg" alt="Image" /></td>
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![Image](image4.jpg)
Paper based device

TB positive serum  TB negative serum  Control (no serum)
Paper based device
Success!

Score 0 = 159.0962
Score 1 = 158.2315
Score 2 = 153.203
Score 3 = 124.912
Score 4 = 147.9861
Score 5 = 149
Score 6 = 150.141
The second stage of the development will have extra controls built into the application to take account of variations in lighting.
Imminent work

The flow through device needs optimizing in conjunction with the phone application

We will then test the known 350 WHO serum samples

Most likely re-optimize

Then test 200 of FINDs blind serum samples
Conclusions

Lipid antigens do detect TB antibodies
We get different responses for different mycobacterial infections
The response is dependant on which lipid antigens are used
We can get a result in under 10 mins, which is readable by eye and/or by phone for under 10$

Needs no external power supply
The response is not affected by BCG and does not appear to give a signal from latent TB
Thanks you for your attention

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