EXPOSING EXPLOSIVES

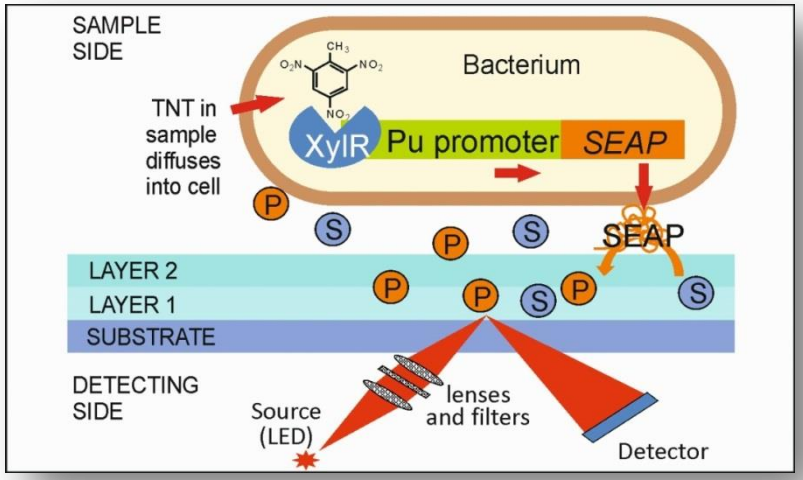
TNT Detection via Innovative Waveguide Sensing

The Problem

The advancement of technologies, in combination with criminal expertise and the availability of explosives increases the potential for terrorists to evade existing UK security systems. Additionally, explosives are present as toxic environmental pollutants on military training sites. There are currently no effective, inexpensive devices that offer high throughput, rapid and specific detection of explosives.

The Concept

The aim of this work is to create a biosensor device for the detection of the explosive TNT. A whole-cell bacterial system will be coupled to an innovative optical waveguide detector to form a device that will enable fast, simple and reliable detection of low levels of TNT.

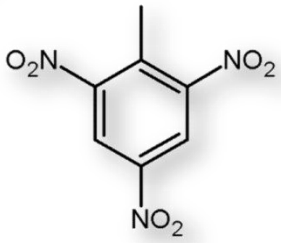


Concept for whole-cell waveguide biosensor

Injection moulded waveguide chip

***p*-xylene**

Trinitrotoluene



The Device

XylR is a transcription factor protein found in the 'oil eating' bacterium *Pseudomonas putida.* When 'activated' XylR can cause the bacterial cell to begin producing various proteins. It has been shown that XylR can be activated by a number of effector substances including xylene and TNT.

We will construct a whole cell biosensor by engineering bacterial cells so that the XylR transcription factor with respond to the presence of TNT by promoting the production of secreted alkaline phosphatase (SEAP) – an enzyme that will be secreted from the cell, and that can catalyse the production of a product outside the cell which will be detected by the optical waveguide sensor.

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[dstl]
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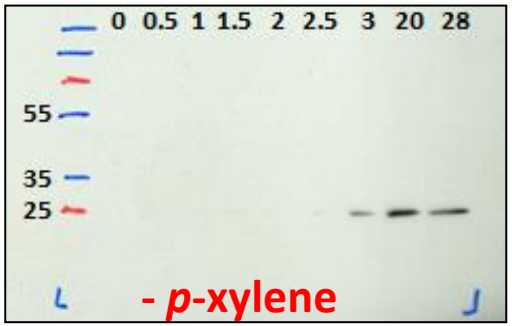
Joe Bennett, Behnam Bastani, Liz Rylott, Nick Goddard and Neil Bruce

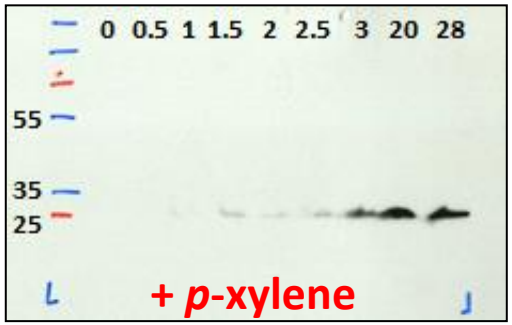
Biosensing Cells…so far!

On our way to engineering the final biosensor device, we have produced bacterial cells that will express a Green Fluorescent Protein (GFP) in response to *p*-xylene. Although we see a background level of GFP when *p*-xylene is absent, we have shown an increased response when it is present at 1 mM concentration.



In the absence of the effector the cells produce detectable levels of GFP after 3 hours.

GFP size ~26.9 kDa

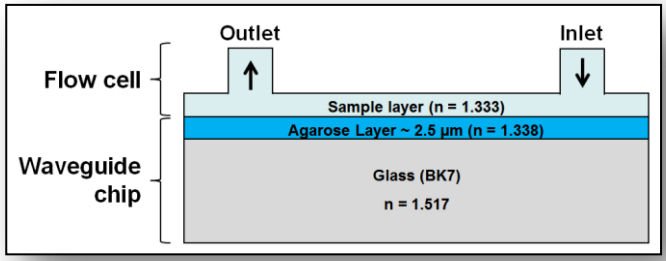


In the presence of the effector cells produce GFP after 1.5 hours.

These figures show western blots probing for GFP.

'Leaky' Waveguide Detector

The waveguide comprises of a thin layer of agarose coated onto glass. At a certain angle, a light beam directed into the waveguide will bounce back and forth along the agarose layer, eventually 'leaking' out to be detected. The intensity of this leaked light is directly affected by the solution in the flow cell.



We have shown that the waveguide can detect ~2 ng mL^{-1} alkaline phosphatase activity – this is ~15x lower than standard techniques.

Future work

Work on the TNT waveguide biosensor is still ongoing. We have shown that the waveguide can detect very low levels of alkaline phosphatase – the enzyme we hope our biosensing cells will secrete in the presence of TNT. We have engineered biosensing cells that produce GFP in response to the effector *p*-xylene, and we are currently moving on to create cells that will secrete alkaline phosphatase enzyme in response to TNT.

For further information: neil.bruce@york.ac.uk

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