# Rapid detection of pathogens in drinking water supplies in rural Malawi — preliminary results

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#### Introduction

Drinking water supplies contaminated with bacteria and viruses cause diarrheal diseases, which kill 1.8 million people a year (WHO, 2016). Sustainable Development Goal 6 highlights the need to address this issue. Currently, pathogen presence is commonly assessed by culturing thermotolerant coliforms (TTCs) (assumed to be *Escherichia coli*) as surrogate indicator organisms, however, this is an intensive and time-consuming laboratory process. Early studies have shown that the use of tryptophan-like fluorescence (TLF) as a real-time indicator of thermotolerant coliforms in the field is promising, through comparison with established laboratory culturing methods. In this study we test the application of TLF in the field in Malawi.



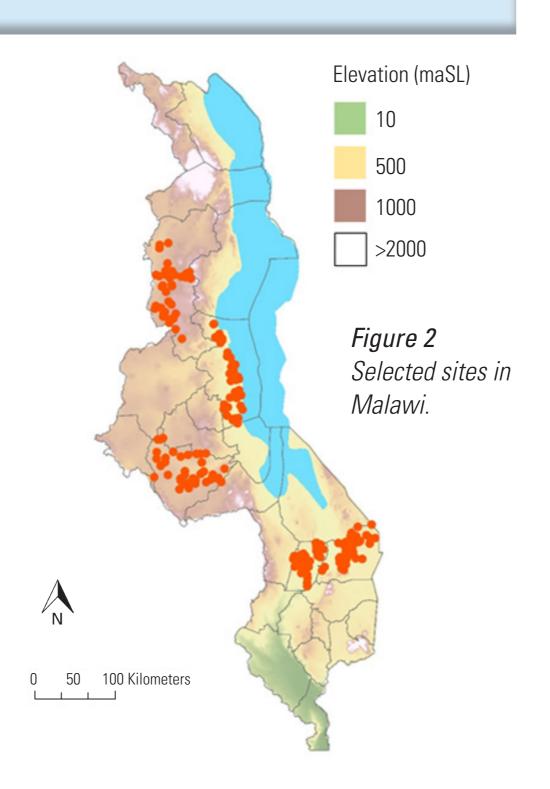




Figure 1 Sources of water in rural Malawi. Left: shallow well; Centre: hand pumped borehole with Afridev pump; Right: tap stand.

# The study

This study investigates the bacterial water quality of drinking water supplies in rural Malawi, at a national scale, employing both TLF and TTC culturing methods. A rigorous hierarchical approach to the sampling (a two stage stratified randomised design) was undertaken, to ensure a representative data set was collected. A total of 183 water sources across five districts were sampled in the dry season to determine the effectiveness of TLF as a rapid screening tool. 162 hand pumped boreholes and 21 alternative sources, including shallow wells and tap stands, were selected to ensure a range of water quality was sampled (Figure 1). Sampling was undertaken in five districts in the 2016 dry season (Figure 2).



## What is Tryptophan-Like Fluorescence?

Using fluorescence to measure water quality parameters is an established approach. Tryptophan is an amino acid found within microbiological cells and fluoresces within the approximate region of 280 nm excitation/350 nm emission wavelength (Figure 3). In surface water and wastewater, 'tryptophan-like fluorescence' (TLF) has been found to correlate with elevated concentrations of nitrate and phosphate originating from sewage inputs due to increased microbial activity (Baker & Inverarity, 2004). More recently, this technology has been applied to groundwater quality (Sorensen et al., 2015). A portable UviLux fluorimeter developed by Chelsea Technologies Group Ltd, UK, was used for this study (Figure 4).

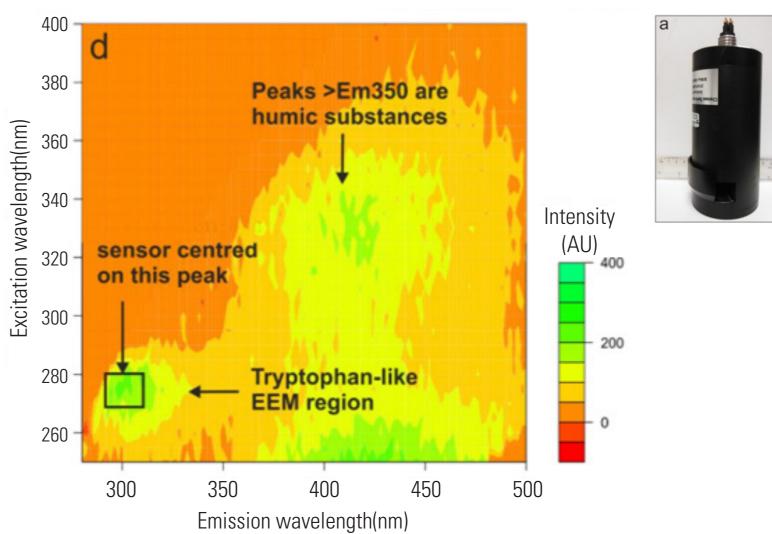


Figure 3 Excitation-emission matrix (EEM) highlighting peaks for TLF and humic substances (NERC, 2017).



WHO Health

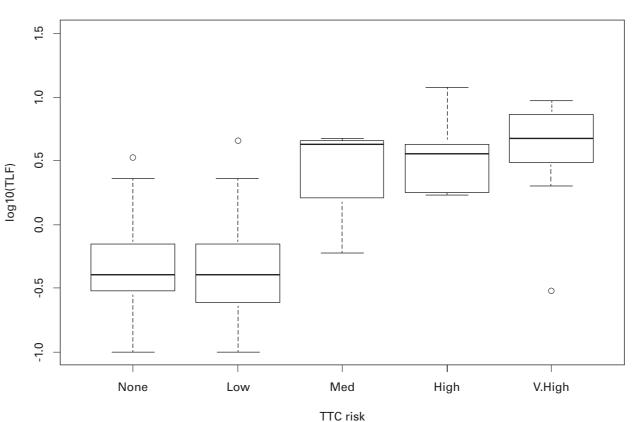
Risk

Figure 4 UviLux TLF probe and use in field (Left: NERC, 2017; Right: Ward, 2017).

No. of TTC

coliforms

### Results



| (cfu/100ml) | Category       |
|-------------|----------------|
| 0           | No risk        |
| 1–9         | Low risk       |
| 10–99       | Medium risk    |
| 100–999     | High Risk      |
| >=1000      | Very high risk |

**Figure 5** Comparison of TLF data with WHO health risk categories for TTC presence.

**Table 1** WHO health risk categories.

- 1. TLF increases with increased TTC presence despite some scatter
- 2. TLF signal differs significantly (P=<0.001) between sites with and without TTC presence. The false positive rate is 20% and false negative rate is 10%. This suggests TLF has potential to screen for contamination but requires some refinement
- 3. TLF detected a significant (P=<0.001) difference between the TTC WHO health risk categories for no and low risk when compared with high and very high risk categories
- 4. Elevated TLF signal at sources with no or low TTC counts could be attributable to the presence of non-culturable cells due to environmental conditions, which means TLF could be a more sensitive indicator of bacterial contamination than TTC culturing
- 5. A high TTC count and low TLF signal at one source was shown to be due to TTC not being *E. coli* (an assumption of the TTC method)

### **Conclusions and further work**

This study concludes that TLF has potential to be developed as a rapid screening tool for identifying contamination in drinking water, however a better understanding of the factors influencing TLF and the inherent variability of the TTC method is needed to reduce false positives and negatives.

Further work includes re-sampling sources in the wet season, where compartment bag tests and Colilert were used to identify *E. coli* contamination specifically and the proportion of TTCs that were *E coli*, in addition to repeat TLF sampling and TTC culturing. Future work is planned to investigate the variability within the TTC method.

WHO, 2016. WHO Drinking-water Fact Sheet. WHO. Available at: http://www.who.int/mediacentre/factsheets/fs391/en/ [Accessed July 8, 2017].

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Sorensen, J.P.R. et al., 2015. In-situ tryptophan-like fluorescence: A real-time indicator of faecal contamination in drinking water supplies. Water Research, 81, pp.38–46.

NERC, 2017. In: Sorensen, 2015. Real-time inference of thermotolerant coliforms in groundwater, 42nd IAH Congress presentation. Available at: http://nora.nerc.ac.uk/512301/1/IAH2015%20E-Poster%20TLF.pdf [Accessed August 23, 2017]

**Contact information**