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Title:

In-Situ detection of microcystin in a pre-lysed freshwater sample using an integrated centrifugal microfluidics platform

Authors & affiliations:

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Abstract: (Your abstract must use **Normal style** and must fit in this box. Your abstract should be no longer than 300 words. The box will 'expand' over 2 pages as you add text/diagrams into it.)

Preparation of Your Abstract

1. The title should be as brief as possible but long enough to indicate clearly the nature of the study. Capitalise the first letter of the first word ONLY (place names excluded). No full stop at the end.
2. Abstracts should state briefly and clearly the purpose, methods, results and conclusions of the work.

Introduction: Clearly state the purpose of the abstract
Methods: Describe your selection of observations or experimental subjects clearly
Results: Present your results in a logical sequence in text, tables and illustrations
Discussion: Emphasize new and important aspects of the study and conclusions that are drawn from them

Introduction:

Cyanobacterial toxins from *Microcystis aeruginosa* predominantly manifest in brackish and fresh water sources and have been shown to promote hepatic disease and cancer [1 - 3]. Autonomous and continuous monitoring of these hepatotoxic cyanobacterial toxins is particularly challenging, and thus critical to environmentally monitor them. Here, we present a portable and cost-effective centrifugal microfluidics-based integrated system for in-situ detection of toxins from *microcystis aeruginosa*. Our unique system includes an in-house developed, and optimised, competitive assay for microcystin toxins, with a top-down LED-photodiode detection system also developed and 3D printed in-house. Preliminary results demonstrate that the system detects cyanobacteria toxins in pre-lysed freshwater at World Health Organisation recommended detection limits of 1ng.ml^{-1} [2].

Methods:

The centrifugal microfluidic-based system, A.K.A. ToxiSense, consists of two components; a multi-layered microfluidic disc for fluidic control and multiplexed assay integration; and a detection system. Also included is a software controlled motor for disc spinning as well as a top-down fluorescent detection technique for *Alexa 430.*

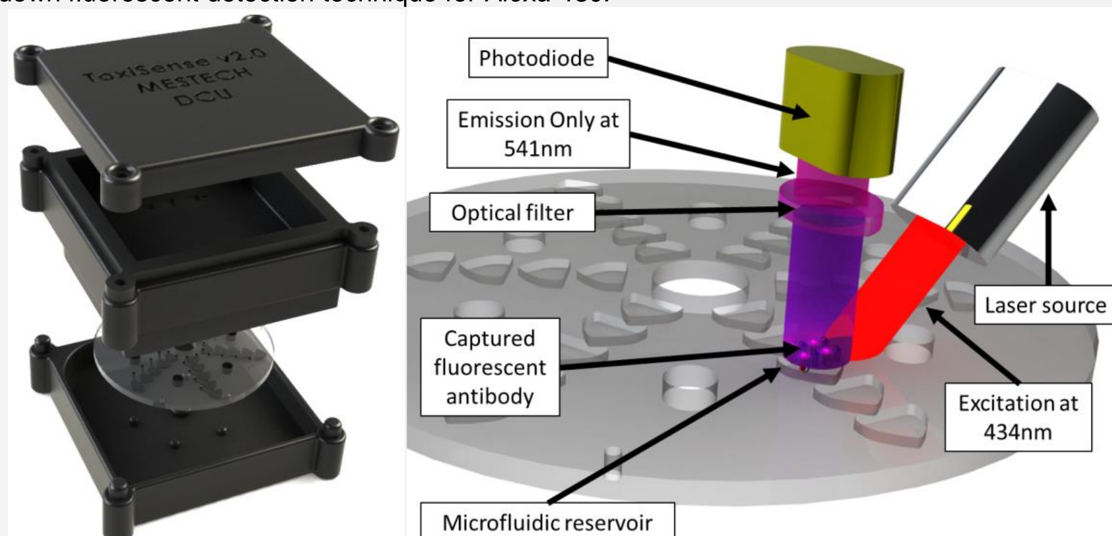

Figure 1: The microfluidic detection system

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The microfluidic disc is manufactured from poly-(methyl-methacrylate) (PMMA) sheets and pressure-sensitive-adhesive (PSA) (ARseal™90880), sourced from Radionics™ and Adhesives Research™ respectively. The Disc consists of five reservoirs, each with separate ventilation systems, in radial alignment connected by microchannels. Each reservoir is a single step in the assay protocol (shown below), with reservoir four and five acting as the control and waste reservoirs, respectively.

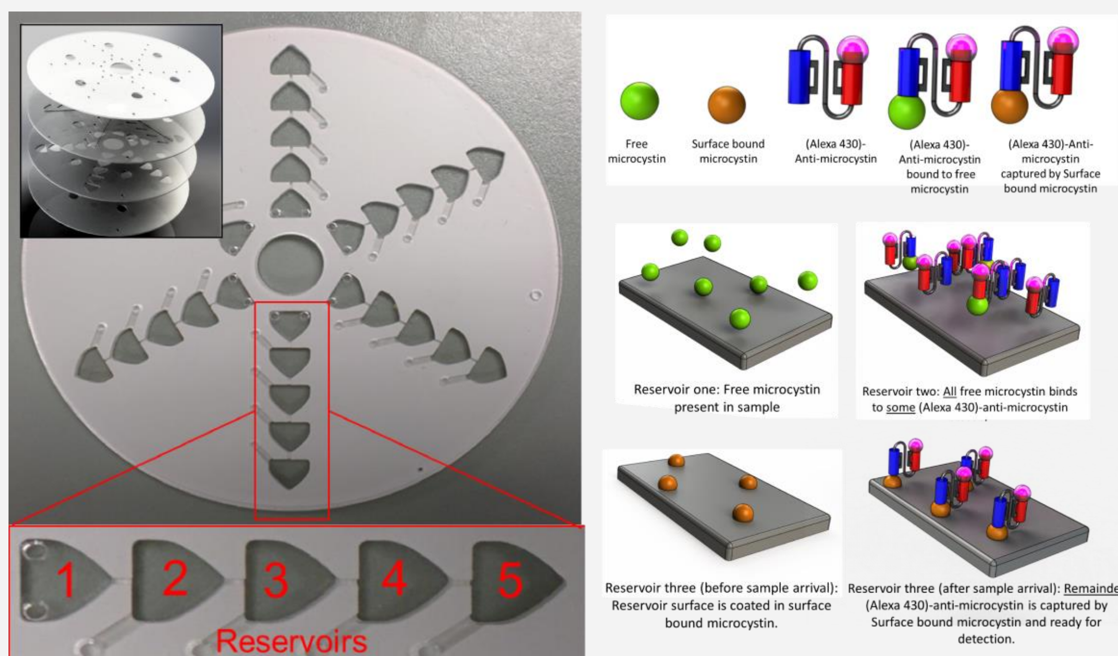

Figure 2: The bio-activated microfluidic disc

Results and Discussion:

Using the ToxiSense platform, the results in Table 1 illustrate that analyte (microcystin) concentration is inversely proportional to fluorescence response. This simple, easy to use system offers a novel approach to in-situ observation of microcystin. The entire system including the assay, microfluidic platform and detection system will continue to be optimised in order to achieve an autonomous and continuous environmental monitoring system.

Table 1: Microcystin competitive assay

Microcystin concentration (ng/mL)	Normalised Relative fluorescence (a.u.)
10000.0	0.8640
1111.1	0.8953
41.2	0.94825
13.7	1.0024
0	1

References

[1] C. MacKintosh, *et al.*, “Cyanobacterial microcystin-LR is a potent and specific inhibitor of protein phosphatases 1 and 2A from both mammals and higher plants,” *FEBS Lett.*, vol. 264, no. 2, pp. 187–192, 1990.

[2] World Health Organization, *Toxic Cyanobacteria in Water: A guide to their public health consequences, monitoring and management*. 1999.

[3] J. M. Rinta-Kanto, *et al.*, “Lake Erie Microcystis: Relationship between microcystin production, dynamics of genotypes and environmental parameters in a large lake,” *Harmful Algae*, vol. 8, no. 5, pp. 665–673, 2009.

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