

# OPTICAL MULTIPLEXING OF MULTIPLE FLUORESCENCE SENSORS FOR COMPACT LAB-ON-A-CHIP SYSTEMS

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## ABSTRACT

Frequency domain multiplexing combined with optical waveguides provides a low cost and efficient approach to optical detection in arrayed microfluidic chips by eliminating detection components and improving acquisition time. A waveguide test chip fabricated in PDMS demonstrates 4:1 multiplexing in the readout by utilizing multilayer waveguides.

**Keywords:** Multiplexing, Optical Detection, Waveguides, Fluorescence Sensor

## 1. INTRODUCTION

Many biochip systems require optical detection at discrete locations, such as flow cytometers, PCR chips, and bioreactors [1, 2, 3]. Although only requiring point detection, these systems usually have high bandwidth requirements, necessary for high-throughput cell sorters or quantitative fluorescence lifetime detection. To provide sensors for truly portable lab-on-a-chip systems, optical detection must be made compact and cheap, utilizing non-imaging optics such as waveguides and reducing off-chip components such as photodetectors and photomultiplier tubes. Many different approaches to waveguide fabrication have been demonstrated for integration with microfluidic systems [1, 4]. In addition, frequency division multiplexing (FDM) has been explored as a method to reduce component costs and increase speed for multichannel fluorescence microscopes [5]. We present a new detection approach combining waveguides with frequency multiplexed fluorescence detection as a low cost and scalable approach for accurate detection of multiple fluorescence signals located within a single chip.

## 2. DESIGN AND FABRICATION

The test chip fabricated is shown in Figure 1. Four individually modulated excitation waveguides are connected to vertical collection waveguides at the

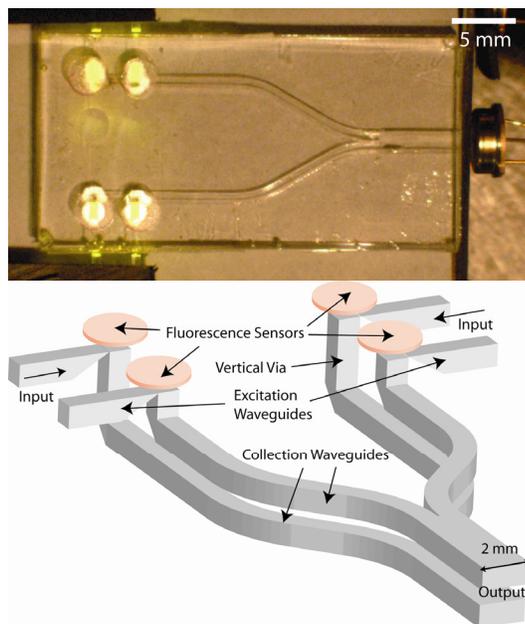


Figure 1. Image and schematic of the waveguide chip. Fluorescence sensors in the fluidic chip are positioned to interface to the detection sites of the waveguide.

optical sensor locations. The collected light is then reflected in plane and combined through a power combiner resulting in all four fluorescence signals converging at a single photodetector. Waveguides with  $1 \times 1 \text{ mm}^2$  cross-sectional areas are fabricated using CNC machining of polycarbonate and vapor polishing, followed by cold injection molding of PDMS into the polycarbonate masters [3]. The waveguide bends of each PDMS layer are coated with 200 nm silver mirrors and the layers are plasma bonded. The resulting hollow channels are filled with NOA71 (Norland Products) and cured under UV illumination. Fabricated  $1 \text{ mm}^2$  waveguides measure a propagation loss of 0.1 dB/cm.

### 3. EXPERIMENTAL

The performance of the system is demonstrated by monitoring the oxidation of sulfite – this reaction consumes oxygen and generates an acidic product [6]. The fluidic test chip above the waveguides consists of a reaction chamber, a waste chamber, and a series of peristaltic mixing tubes to provide oxygen for an oxidation reaction. Two detection sites within the reaction chamber contain a pH sensitive (Presens) fluorophor with an excitation modulated at 44 kHz and an oxygen sensitive dye (PtOEP [7]) modulated at 5.08 kHz. In addition to signals from the pH and oxygen sensors, two waveguide ports are used to monitor the fluorescence from CdSe nanoparticles (or quantum dots), also modulated at frequencies close to the chemical sensor modulation frequencies.

### 4. RESULTS

As shown in Figure 2, the overlapping spectra of the fluorescent sensors make simultaneous measurements difficult in wavelength domain. However, with excitations modulated at different frequencies as shown in Figure 3, all four signals are clearly distinguishable from one another. FDM is able to extract data from the sensors

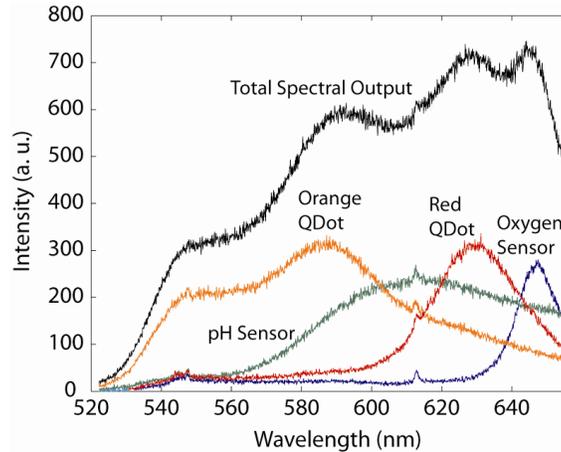


Figure 2. Emission spectra of the waveguide output incident on the photodetector. Oxygen and pH are still distinguishable with frequency multiplexing under large spectral interference.

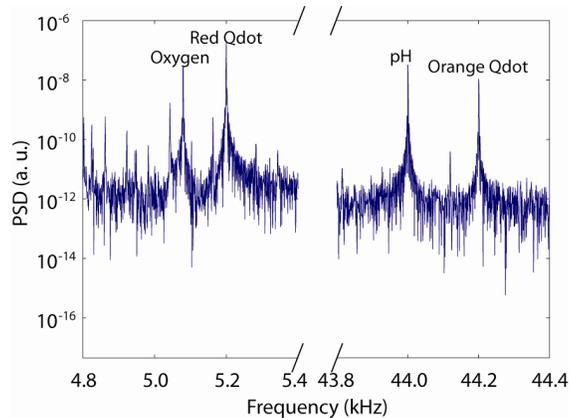


Figure 3. Power spectral density of the output photodetector clearly indicating the modulation frequencies of each fluorophor.

despite the large overlap in emission between different fluorophors. As seen in Figure 4, the oxidation of sulfite consumes the available oxygen and steadily reduces the pH in the chamber. This reaction proceeds until the sulfite begins to deplete, indicated by the gradual rise in oxygen concentration.

## 5. CONCLUSIONS

Frequency division multiplexing combined with optical waveguides provides an approach for addressing simultaneous detection in microfluidic arrays. In this work we have demonstrated a 4x multiplexed fluorescence detector capable of both intensity based and lifetime based fluorescence detection.

## ACKNOWLEDGEMENTS

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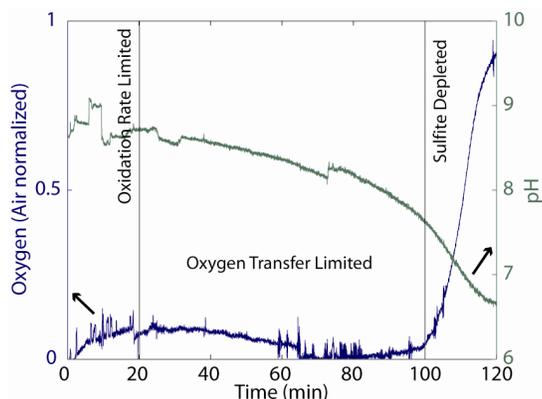


Figure 4. Measured oxygen and pH of a Sodium Sulfite oxidation reaction using the integrated waveguide FDM sensor.