GENERATION AND DETECTION OF LAMINAR FLOW WITH LATERALLY-VARYING OXYGEN CONCENTRATION LEVELS

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ABSTRACT

This paper describes the generation and detection of laminar flow with laterally-varying oxygen concentration levels via luminescence-based optical oxygen sensors in a microfluidic device. Analysis of the measured oxygen concentration profiles was used to determine the diffusion coefficient of oxygen in water.

KEYWORDS: Bioreactor, Dissolved O_2 Sensor, Laminar Flow, O_2 Diffusion

INTRODUCTION

Dissolved oxygen (DO) is an important parameter in biomedical and cell-culture applications [1]; various studies have found cell survival and function to be intimately linked to oxygen concentration [2,3]. Recently we have demonstrated a novel process for the fabrication of platinum(II)octaethylporphyrin ketone/ polystyrene (PtOEPK/PS) oxygen sensors based on spin-coating, polydimethyl-siloxane (PDMS) soft-lithography and reactive ion etching (RIE) [4]. In this study the process is used to integrate sensors into a microfluidic cell-culture device for in-situ measurement of DO concentration profiles.

FABRICATION

The process of sensor fabrication and integration is shown in Figure 1. A stamp of the desired sensor patterns is replicated in PDMS from a SU-8 resist master, brought into conformal contact with the spin-coated sensor layer and transferred into the layer by RIE in oxygen plasma. Figure 2 shows an example of a sensor pattern.

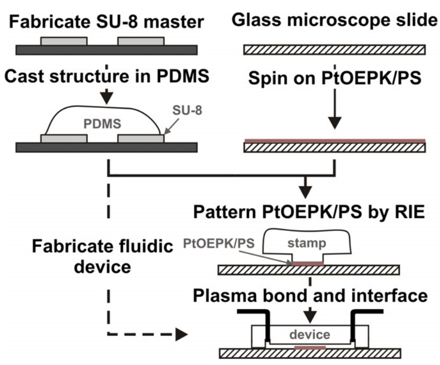

Figure 1. Schematic of the sensor and device fabrication process

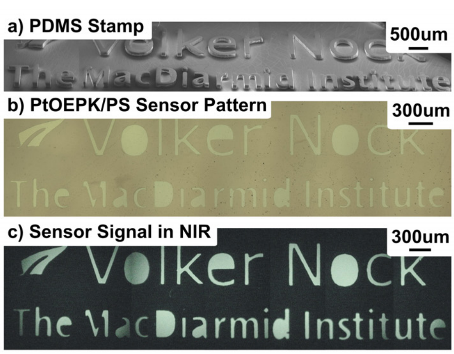

Figure 2. Test pattern replicated using the fabrication process

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Parallel to sensor fabrication a PDMS microfluidic device with integrated gas exchanger was replicated from a second SU-8 master. This microfluidic device was bonded to the substrate containing the sensor pattern using surface activation in oxygen plasma. Figure 3 shows a photograph of the final device with two external gas exchangers (a) and a SEM of the PDMS membrane providing oxygenation by gas diffusion into the liquid channel (b).

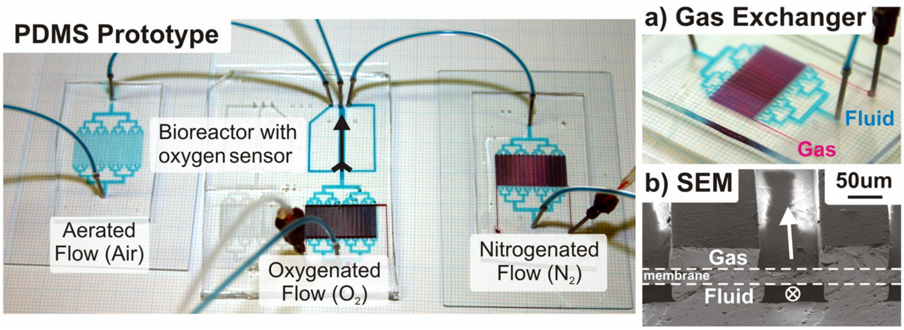

Figure 3. Prototype device providing parallel laminar flow of different DO concentrations. Integrated gas exchangers (a) provide for oxygenation of a liquid via gas diffusion through a PDMS membrane (b). A fluorescence-based oxygen sensor in the bioreactor allows for in-situ monitoring of the DO concentration

EXPERIMENTAL

Oxygen measurement was performed using a fluorescence microscope in combination with a digital camera for image acquisition. The change of sensor intensity was analyzed using the image processing module of Matlab and a pre-recorded Stern-Volmer calibration curve, as described in detail in [4]. Three gas exchangers and a syringe pump were used to provide parallel flows of different oxygen concentrations. These flows were combined in a central parallel-plate reactor chamber to form laminar flow with laterally varying DO levels.

RESULTS AND DISCUSSION

The integrated gas exchangers were measured to provide a maximum DO concentration of 34 ppm in de-ionized (DI) water. At a flow rate of 1.0 mL/min the 500 nm thick PtOEPK/PS sensor layer yielded an intensity ratio of $I_0/I_{100} = 5.9$ for exposure to 0% and 100% DO. Three flows, each of 0.1 mL/min and 0, 34 and 8.6 ppm DO, respectively, were combined in the rectangular parallel-plate reactor chamber via the separate inlets.

Figure 4(a) shows fluorescence intensity images recorded at three locations along the length of the chamber, visualizing the different oxygen concentrations of the flow streams. Individual streams remain separated along the full length of the reactor due to laminar flow conditions. To demonstrate the applicability of the device for use as lab-on-a-chip the diffusion coefficient of oxygen in water was measured. Quantitative analysis of the fluorescence images at the inlet and outlet is shown in Figure 4(b). Comparison of the change in slope between the two concentration profiles using diffusion theory yields a diffusion coefficient of oxygen in water of 2.56×10^{-5} cm^2/s at 37°C, which agrees well with literature [5].

CONCLUSIONS

We have shown the generation and detection of laminar flow with varying levels of oxygen concentration inside a PDMS-based microfluidic device. Analysis of the measured oxygen concentration profiles was used to determine the diffusion coefficient of oxygen in water. The presented chip provides a novel tool for the parallelization of DO concentration-dependent assays, bioreactor and tissue engineering applications.

ACKNOWLEDGEMENTS

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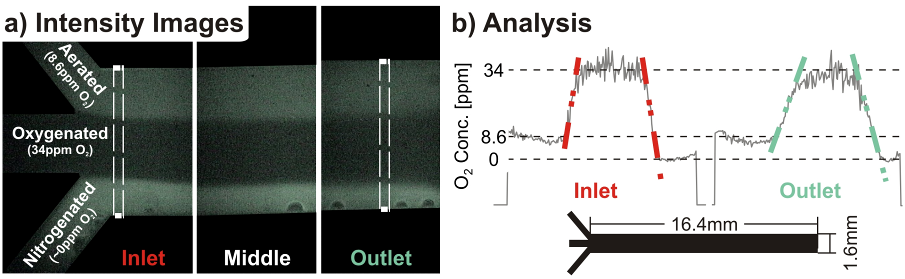

Figure 4. (a) Fluorescence images of the optical detection of laterally-varying oxygen concentration inside the parallel-plate bioreactor chamber. (b) The analysis of the slope of the concentration profiles yields a diffusion coefficient for O_2 in DI water of $2.56 \times 10^{-5} cm^2/s$

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