

A SINGLE POINT EVANESCENT WAVE PROBE FOR ON-CHIP REFRACTIVE INDEX DETECTION

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Abstract

A novel method to detect refractive index variations within microchannel environments using an evanescent wave probe is described. Experiments, with and without a fluorescent buffer, demonstrate that 10 mM sucrose plugs can be detected. This method is also applied to the detection of sucrose plugs flowing hydrodynamically through microchannels and for chip-based electrophoretic separations of saccharide mixtures.

Keywords: Refractive index, evanescent wave, microchip

1. Introduction

Although fluorescence spectroscopy has tended to be the detection method of choice for chip-based analysis, the majority of molecules are non-fluorescent. Therefore, several alternative approaches to ‘on-chip’ detection have been demonstrated. The potential of refractive index (RI) detection (universal, non-destructive) for chip-based analytical systems has been investigated [1,2]. We present a simple method to detect RI changes within microfluidic channels based on the principles of total internal reflection (TIR).

2. Theory

Snell’s Law can describe the behavior of electromagnetic radiation at an interface.

$$n_1 \sin \theta_1 = n_2 \sin \theta_2$$

where n denotes the RI and θ is the angle at which the radiation enters or leaves the interface. If n_1 is greater than n_2 , then there exists for θ_1 an angle, called the critical angle, where $\sin^{-1}(n_2/n_1)$ is $\pi/2$. If θ_1 exceeds this condition, then all incident radiation is reflected at the interface. However, solving Maxwell’s equations for this system shows that an evanescent wave (EW) exists in medium 2 in the vicinity of the boundary and is directly affected by the difference in the refractive indices of the two media. This variation is the basis of the proposed sensing mechanism.

3. Experimental

Details of the optical set-up have been described elsewhere [3]. RI measurements using the EW induced fluorescence system (EWIFS) were either in a Teflon flow cell or microfluidic channel (200 x 80 μm) were filled with the desired sucrose solution. The incidence angle was scanned from 75 to 65 degrees and the fluorescence measured. For

repeat injection experiments, the chip was connected to a Rheodyne injection valve with a 5.44 μL sample loop using 152 μm ID PTFE tubing. Solutions were pumped through the chip using a Harvard Apparatus pump at a rate of 10 $\mu\text{L}/\text{min}$.

4. Results and Discussion

Experiments to measure RI variations with and without (Figure 1) a fluorescein solution gave results in close agreement with literature and standard refractometer values. Repeat injections of sucrose with (Figure 2) and without fluorescein were passed through a microfabricated chip. As sucrose destroys the formation of the EW, it is detected due to a large increase in fluorescence from the bulk solution. The same technique was used to detect electrophoretically separated saccharide mixtures. By setting the incidence angle of the laser at a defined angle, hence defining the RI of interest, injections can be made discriminating against lower RI and detecting those of higher RI.

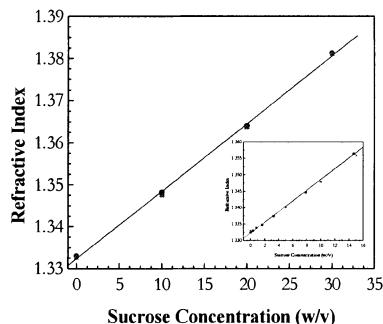


Figure 1

RI measurement of various sucrose conc.: literature (-), refractometer (●), EWIFS (■). The inset shows values from 0 to 500 mM.

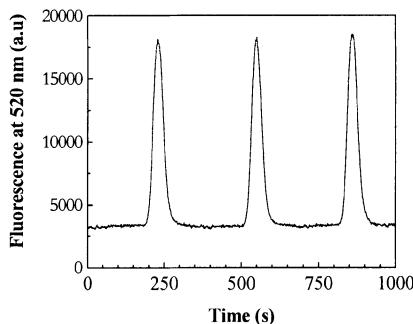


Figure 2

Repeat injections using a Rheodyne valve of 25 mM sucrose in 2 μM fluorescein through a microfluidic channel. The running buffer was 2 μM fluorescein.

5. Conclusions

It has been shown that a single point evanescent probe can be used to detect RI changes on chip. This simple technique provides a first step towards a universal, small-volume detector for chip-based separation systems.

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References

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