

Ultra-Sensitive Optical Biosensor Based on Whispering Gallery Modes: The Effect of Buffer Solutions Refractive Index on Their Sensitivity and Performance

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ABSTRACT

Background: Whispering gallery modes (WGM) biosensors are ultrasensitive systems that can measure amount of adsorbed layer onto the micro-cavity surface. They have many applications including protein, peptide growth, DNA and bacteria detection, molecular properties measurements and specific interaction and drug table recognitions due to their high sensitivity, compact size and label free sensing mechanism.

Objective: In this paper we investigate the effect of buffer solution on detection of specific biomolecules in WGM biosensors through its refractive index change.

Method: The propagation of electromagnetic waves in a dielectric microsphere is analyzed by solving Maxwell's equations through proper boundary condition to find a concise relation for micro-cavity resonance shift.

Results: Analysis of the buffer solution's refractive index effects on detection of BSA by WGM biosensors are presented and it was shown that even a very small change in the refractive index of buffer solution can affect the biosensor wavelength shift and the sensitivity of biosensors.

Conclusion: This study opens up a discussion in biosensor sensitivity based on true and reliable performance of the buffer solution through its accurate determination of refractive index and behavior. To avoid expensive methods of enhancing sensitivity, one can improve the sensitivity of WGM biosensor to some extent, by means of using proper buffer solution.

Keywords

Biosensors, Whispering Gallery Mode, Wavelength Shift, Sensitivity, Microsphere, Refractive Index, Buffer Solution

Introduction

High potential application of miniature detectors and measuring tools in various fields such as pharmaceutical, medical, and chemical industries results in development of sophisticated biosensor systems [1]. Thanks to properties such as immunity to electromagnetic noise, ability of remote sensing, short response time and high sensitivity, they have become practical devices which have much preference over tedious spectroscopic techniques.

Whispering gallery mode (WGM) label free optical biosensors have been designed and successfully worked out in the field of bio-molecular sensing [2-11]. The origin of WGMs returns to light propagation near the surface of a circular dielectric micro-cavity by total internal reflections

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(TIRs) [12]. High Q factor, very small mode volume and narrow resonant spectral line of optical micro-cavity are important for designing high sensitive, accurate sensor systems. High sensitive bio-molecular sensing system based on WGM was proposed by Vollmer et al [8]. Single molecule detection by WGM biosensor was also reported [2, 17]. Different biological materials including BSA, DNA, bacteriorhodopsin, trypsin, mercury and peptide growth process have also been studied by WGM sensors [4-8, 11, 22]. Moreover, very small volume of sample was also detected by WGM spectrum analysis [9]. A resolution on the order of RIU (refractive index unit) for bulk RI (refractive index) variation was also reported [3]. Although the Q factor of microspheres decreases in aqueous solution, sensing process in liquid environment, has been performed with the sensitivity as high as surface plasmon resonance detection method [4]. Mechanical, temperature and gas detection sensors have also been developed based on micro-cavity resonance modes [13-16].

The principles of WGM biosensors operation can be described based on the shift of resonance wavelength against any alteration of the microsphere size or any change in the optical properties of the surrounding medium. Therefore adhesion of bio-molecule on the surface of microsphere leads to the change of the microsphere size and the optical properties of the surrounding medium and results in the shifts of the resonance wavelength. Previous analyses of these wavelength shifts provide valuable information about the quantitative and qualitative aspect of the bio-molecule. Therefore, various parameters such as, microsphere size, light coupling efficiency to microsphere and the refractive index of materials are vital and important parameters in WGM biosensors. As the beforehand experimental methods to optimize these parameters are costly, precise modeling and simulation can help in understanding the behavior and performance of such delicate

systems.

As the effect of buffer solution is so important in achieving accurate and reliable data, many works can be found in the literature that devoted to detailed consideration of solution preparation, knowing various ambient factors [18-24] and the dependency of optical absorption spectrum to solvent RI [19]. In this regard, the way that concentration of buffer solution can dramatically affect the detection of tiny amount of proteins was also reported [20]. In this paper the effect of bio-sensor buffer solution on the WGM sensor operation is studied. As the refractive index of buffer solution undergoes change under variation of ambient parameters such as temperature, pressure and impurities, the alteration of buffer's RI got the main key in sensor performance and this in turn can reflect the ambient factor changes. One of the most important parameters in working stages of sensors is their sensitivity and responsivity to different real and noisy signals. Bad choice of buffer solutions not only increase costly demand for high sensitive devices but also can fool the experimenter in reporting high value data from reliability, reproducibility and precision point of view. Therefore, this work somehow identifies PBS buffer solution usually employed in WGM biosensors for accurate determination of various proteins adsorbed in tiny amounts on the circular-shape micro-cavity. It does show that the sensor should contain proper buffer solution and the effect of those solutions should be rigorously taken into account through their calibration curve for better quantitative measurements of proteins. In this way, if proper buffer solution is selected, even low sensitive WGM sensor can provide valuable data.

After a brief review of a spherical micro-cavity's resonance modes, we investigate the WGM resonance response to the surrounding RI change. Taking into account the small variation of solvent RI, which might occur due to ambient factors such as, pressure or

temperature fluctuations, we calculate the corrected value to micro-cavity resonance shift. Since these biosensors are very sensitive to very small change of refractive index, it is shown that the change of solution refractive index can't be ignored in WGM biosensors and should be considered in design and construction of biosensors.

Theoretical formulation

The propagation of electromagnetic waves in a dielectric microsphere is described by Maxwell's equations: [22]

$$\nabla \cdot E = \frac{\rho}{\epsilon}; \quad \nabla \times E = -\mu \frac{\partial H}{\partial t}; \quad (1)$$

$$\nabla \cdot H = 0; \quad \nabla \times H = \epsilon \frac{\partial E}{\partial t} + J$$

where E, H, μ , ϵ , J and ρ are electric field, magnetic field, magnetic permeability, electric permittivity, current density, and free electric charge, respectively.

Considering time-harmonic TE wave:

$$E(r, t) = E_z(x, y) e^{i\omega t} \vec{e}_z \quad (2)$$

We can derive the following equation for E:

$$\frac{1}{\mu} \nabla^2 E + \omega^2 \epsilon E = 0 \quad (3)$$

Eq.(3) is the wave equation which can be used for finding the exact solution of WGM electric fields. The solution of Eq.(3) both in inner and outer regions of micro-cavity, with proper boundary conditions at the interface yields the resonance wavelength and WGM fields. Fig.(1) shows the first radial TE₂₁ whispering gallery mode.

Sensor performance

The key element in sensor performance is the change of the RI surrounding medium that is if RI is changed by δn_s as:

$$n_s = n_{0s} + \delta n_s \quad (4)$$

then a remarkable shift in WGM spectra will

be appeared. Fig. (2), clearly shows such a frequency shift when $\delta n_s = 0.1$. The frequency shift can also occur by any changes of micro-cavity size. When analytes adhered to the micro-cavity surface, the micro-cavity size is changed which would lead to a shift of the resonance frequency. The dependence of WGM resonance wavelength to the molecular weight of analyte has also been demonstrated for protein detection [18].

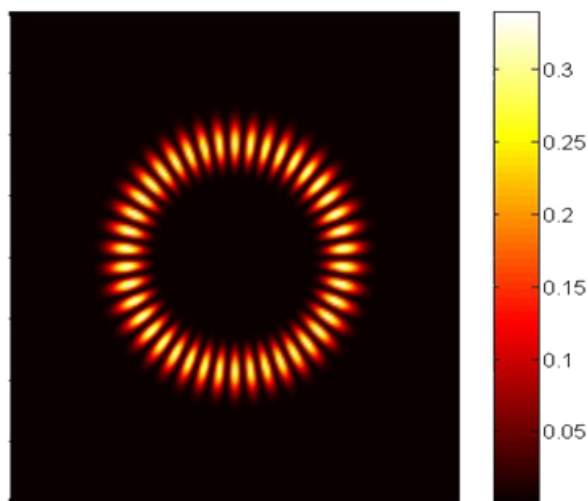


Figure 1: Whispering gallery mode electric field TE₂₁ (First radial mode)

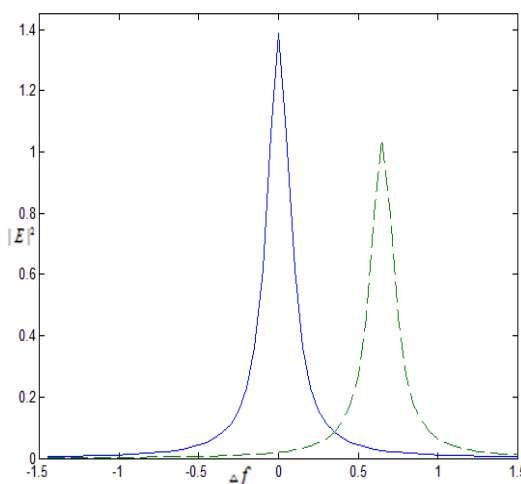


Figure 2: Electromagnetic spectrum for different refractive indices of surrounding medium: $n_s = 1.15$ (solid line), $n_s = 1.25$ ($\delta n_s = 0.1$) (dashed line)

Results and discussions

Taking into account that the refractive index of protein solution is buffer dependent, the surrounding refractive index can be written as [21]:

$$n_s = n_c + \frac{dn}{dc} c_A \quad (5)$$

where c_A is concentration of adsorbed molecules, and n_c is refractive index of buffer solution. dn/dc is the RI increment due to adsorbate. The dependence of buffer solution RI to the temperature and concentration were also investigated [20]. Considering these relations we now investigate the shift of WGM for small variations of buffer solution refractive index. Using numerical methods we calculate the resonance wavelength shift of WGM as a function of molecular weight (Fig.(3)) and adsorbed layer thickness (Fig.(4)). Following

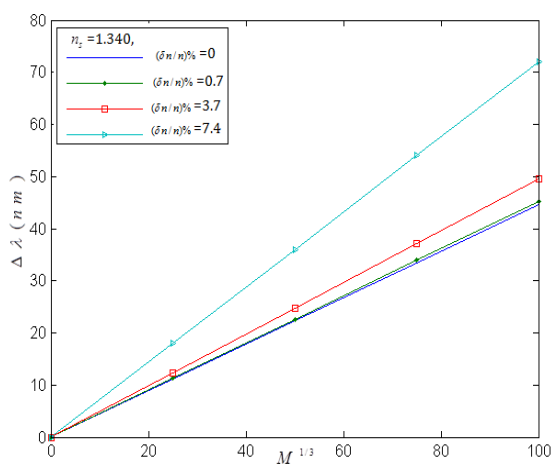


Figure 3: Resonance wavelength shift as function of protein molecular weight for BSA detection for different buffer RI variations

protein for different values of buffer's RI changed. This figure too confirms that low-thickness detection can be done with higher variation of buffers' RI and lower sensitive sensor at least for thickness determination in the range 0-25 nm.

parameters[25] for detection of BSA (bovine serum albumin) dissolved in PBS (phosphate buffered saline) have been used in our calculation: $n_c = n_{pbs} = 1.34$, $n_p = 1.5$, $\rho_p = 1.37(g/cm^3)$, and 0.34 for volume fraction of protein. Fig.(3) shows the wavelength shift of the WGM biosensors versus BSA molecular weight. In this figure, the inset shows one single buffer solution when undergoes different RI variation. As the figures show, one can shift the slope of the curves toward higher wavelength shift for better detection of BSA. One can conclude that at least in the range of $M^{1/3} = 0-100$ the wavelength shift linearly depends on $M^{1/3}$. In this way by rigorous change of the buffer's RI, much lower-sensitive WGM biosensor can be employed. Fig.(4) is also depicting $\Delta\lambda$ versus thickness of the

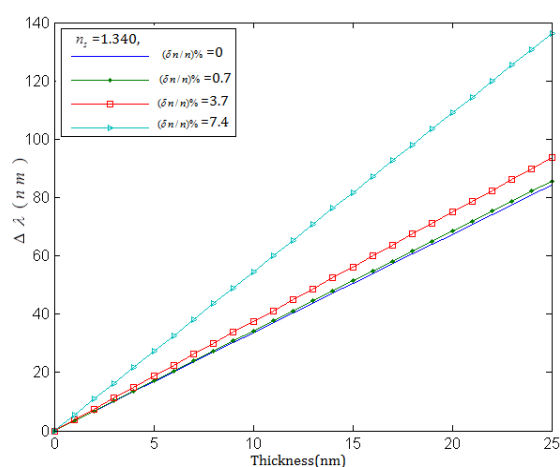


Figure 4: Resonance wavelength shift as function of adsorbed layer thickness for different buffer RI variations.

Conclusion

Ultrasensitive WGM biosensors operation based on the response of resonance modes of micro-cavity to surrounding RI changes which occurs when biomolecules adhere onto micro-cavity surface and interact with the evanescent field of WGM have been studied. Usually in

addition to target molecules, the micro-cavity surrounding medium contains some buffer solution. Therefore optical properties of buffer solution can affect these sensors' operation.

This paper is devoted to the investigation of the buffer solution's refractive index effects on detection of BSA by WGM biosensors and show that even a very small change in the refractive index of buffer solution can affect the biosensor wavelength shift and the sensitivity of biosensors. This work propose that proper choice of buffer solutions where their explicit effects on the RI variation of the whole medium is known can yield valuable and accurate determination of BSA dissolved in PBS without going for costly high sensitive WGM sensors.

Conflict of Interests

None declared.

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