Fluorescence Amplification using Photonic Crystal Slabs

N. Ganesh\textsuperscript{3}, P.C. Mathias\textsuperscript{2}, W. Zhang\textsuperscript{3}, and B.T. Cunningham\textsuperscript{1}

Nano Sensors Group
University of Illinois at Urbana-Champaign

\textsuperscript{1}Departments of Electrical and Computer Engineering, \textsuperscript{2}Biomedical Engineering, and \textsuperscript{3}Material Science and Engineering Urbana, IL

bcunning@illinois.edu

Abstract—Detection of biomolecules and cells through the use of fluorescent tags is the most widely used method in gene expression analysis, biomarker-based disease diagnosis, and cell imaging. We demonstrate the ability to amplify fluorescent output using a photonic crystal (PC) surface, thereby boosting detection sensitivity by orders of magnitude, and apply the approach to several life science applications. We achieve fluorescence amplification by two distinct mechanisms: First, intense optical near fields that develop on the surfaces of a PC slab can be designed to coincide with the absorption wavelength of a fluorophore, resulting in enhanced excitation compared to the same fluorophore excited on an optically inactive surface. Second, PC resonances can be designed to coincide with the fluorophore emission wavelength, enabling enhanced extraction of light. The all-dielectric PC structure offers several important advantages with respect to metal-enhanced fluorescence using surface plasmons.

I. INTRODUCTION

While metal-enhanced fluorescence on the surface of thin films, gratings, and suspended nanoparticles have been demonstrated as an effective means for increasing fluorescence assay sensitivity, fundamental limits due to quenching and the low Q-factor of plasmon resonances have limited their performance. In contrast, we show that PC structures with high Q-factor and lack of fluorescence quenching can provide a large enhanced fluorescence excitation effect. We have characterized the spatial dependence of the enhanced excitation effect using PC slabs (Fig. 1, for example) produced on large surface areas by a nanoreplica molding process[1-3]. This is done by mapping the fluorescence enhancement factor for a fluorophore as a function of its distance above the device surface and comparing the results to numerical calculations (Fig. 2).

II. ENHANCED EXCITATION AND EXTRACTION

A. Guided Mode Resonance

Since their discovery by Wood\textsuperscript{[4]} in 1902 and subsequent analysis\textsuperscript{[5-8]}, anomalies in periodically modulated structures have attracted much attention. Wang and Magnusson studied\textsuperscript{[9, 10]} in great detail resonant anomalies in waveguide-gratings and showed that structures with a sub-wavelength modulation in refractive index along one-dimension can function as filters that produce complete exchange of energy between forward and backward propagating diffracted waves, with smooth line shapes and arbitrarily low linewidths\textsuperscript{[11]}. This anomalous resonant phenomenon (termed guided-mode resonance) arises due to the introduced periodicity which allows phase-matching of externally incident radiation into modes that can be re-radiated into free-space. Due to the fact that these modes possess finite lifetimes within such structures, they are referred to as ‘leaky eigenmodes’ of the structures. More recently, guided-mode resonances have been studied and demonstrated in crossed gratings\textsuperscript{[12-14]} or two-dimensional (2D) PC slabs\textsuperscript{[15-18]}. The leaky nature of these modes has been successfully exploited towards the development of
light emitting diodes (LEDs) with improved extraction efficiency[19-21], biosensors[22] and vertically emitting lasers[23]. In this report, we demonstrate that the external excitation of the leaky modes gives rise to the formation of high intensity near-fields which serves to efficiently excite the organic fluorescent dye molecules and semiconductor quantum dots (QD’s), and the coupling of fluorescent emission to leaky modes overlapping the emission spectrum serves as an effective mechanism to extract this enhanced emission. Such a doubly resonant scheme to enhance fluorescent emission has not been until now demonstrated.

B. Enhanced Excitation

When externally incident light interacts with periodically modulated structures in the sub-wavelength regime (for e.g. PCs), only the 0th order forward and backward diffracted waves can propagate. The periodicity however, also allows for phase-matching of higher (evanescent) orders to localized leaky modes supported by the PC. Once excited, the leaky modes, defined by a complex propagation constant, possesses a finite lifetime as they are leaked out both in the forward (transmitted) and backward (specular) directions. The backward re-radiated waves are in phase and constructively interfere with the 0th backward diffracted order while the forward re-radiated waves are out of phase with the 0th forward diffracted order by \( p \) radians[24], causing destructive interference and consequently resulting in zero transmission. Thus the external excitation of the leaky modes is associated with a 100% reflection phenomenon for the resonant wavelength, assuming a defect-free, lossless system. Since the excited leaky modes are radiative but localized in space during their finite lifetimes, they can be engineered to have very high energy density within regions of the PC at resonance. The magnitude of this energy density is directly related to the resonant mode lifetime or Q-factor of resonance, which in turn can be controlled by adjusting the device parameters. Therefore, the intensity of emission of fluorescent species (that are absorptive at the resonant wavelengths) can be greatly enhanced by placing them in proximity to regions where the resonant modes concentrate most of their energy.

C. Enhanced Extraction

Concurrently, the existence of leaky modes that overlap the fluorescence emission spectrum opens up additional pathways for the emitted light to escape into free-space. Besides direct emission, the fluorescence can couple to the overlapping leaky modes and Bragg scatter out of the structure, thereby greatly reducing the amount of light trapped as guided-modes, in comparison to an unpatterned substrate[20]. If the dispersion of these overlapping emission leaky modes is close to the G-point band-edge i.e. \( K_0 \) (magnitude of in-plane wave vector) \( \sim 0 \), a significant amount of the emitted light will be extracted within small angles with the vertical. It can thus be appreciated that enhancement of fluorescence can be achieved in two steps: enhanced excitation and enhanced extraction.

Enhancing the output of fluorescent species is highly desirable for a wide range of biological applications including DNA sequencing, gene expression, single molecule detection and high resolution cell imaging. An extensive literature exists on the effects of metals on fluorophores, and on the use of a variety of metal surface configurations (planar, corrugated, colloidal) or metallic particle types (spheres, shells, boxes, and triangles to name just a few) that involve plasmonic resonances to enhance fluorescence. A relatively smaller variety of dielectric structures, such as waveguides and optical resonators have been exploited for this purpose. Depending on distance and geometry, metal surfaces can cause increases in either quenching or enhancement of fluorescence. The effects of metallic surfaces on fluorophores are due to at least three mechanisms: (1) Energy transfer quenching to the metal via resonant energy transfer (RET), and thus lower fluorescent intensity. (2) Increase in fluorescent intensity due to the metal amplifying the incident field, where the enhancement is due to the metal particles’ ability to concentrate the local excitation intensity, and (3) An Increase in the intrinsic radiative decay rate of the fluorophore by providing an environment with enhanced photonic mode density resulting in enhancement of quantum efficiency. With a wide variety of surface and structure configurations, plasmon-controlled fluorescence will have an important impact in many fields of biological science. However, surface plasmons present some inherent characteristics that are not ultimately the most desirable ones for enhanced fluorescence. First, fluorescence quenching by the metal surface results in substantial loss of enhancement for any fluorophore within...
small distances from the surface – the same region where the metal enhanced near-fields have their greatest intensity. The quenching may be mitigated by the incorporation of spacer layers, at the cost of lowered enhancement factors. Second, the spectral width of surface plasmon resonances is generally large (~100 nm) due to the lossy nature of metals–essentially implying a resonance with a very low quality factor (Q-factor). As an optical resonator, low Q-factor results in relatively low stored electric field intensity, as energy is quickly dissipated in the metal.

III. RESULTS

Experiments show that the decay of the amplification factor is exponential as a function of distance (Fig. 3) from the device surface and excellent agreement with the numerically calculated exponential decay length is obtained. In contrast to approaches involving surface plasmons on metal surfaces, we find that the maximum amplification is obtained when the fluorophore is closest to the photonic crystal surface due to the absence of quenching at short distances. We also show that the decay length is largely controlled by the wavelength of light resonant in the device. We have likewise characterized the sensitivity gains achievable using PC surfaces to efficiently couple fluorescence emission from molecular dyes and semiconductor quantum dots. We show that the existence of leaky modes that overlap the fluorescence emission spectrum opens up additional pathways for the emitted light to escape into free-space. Besides direct emission, the fluorescence can couple to the overlapping leaky modes and Bragg scatter out of the structure, thereby greatly reducing the amount of light trapped as guided-modes, in comparison to an unpatterned substrate. If the dispersion of these overlapping emission leaky modes is close to the Γ-point band-edge i.e. $K_\parallel$ (magnitude of in-plane wave vector) ~ 0, a significant amount of the emitted light will be extracted within small angles with the vertical, where the light can be captured with a detector. Fig. 4 shows the collected fluorescence emission spectra from quantum dot fluorophores within 1D linear, 2D square, and 2D hexagonal photonic crystal slabs with extraction resonances tuned to the quantum dot emission wavelength compared with the detection of the same quantum dots on an unpatterned glass surface. Combining the enhanced excitation and extraction effects together results in multiplication of the enhancement factors, leading to 500x sensitivity gains for Cy-5 labeled microarray spots (Fig. 5).

IV. CONCLUSIONS

Photonic crystal surfaces represent a class of devices with resonant optical properties that can be exploited for enhancing the detection sensitivity of a broad class of fluorescence-based biochemical assays. The dielectric-based structure does not quench fluorescence, as is the case with metal-enhanced fluorescence based on surface plasmons. Because photonic crystal surfaces can be fabricated over large areas by nanoreplica molding techniques, they can be incorporated into single-use disposable assay formats such as microtiter plates and microarray slides. Many important applications in life science research, including gene expression microarrays, protein biomarker diagnostic tests, and cell imaging, are expected to benefit from photonic crystal fluorescence enhancement.

Figure 3. (a) Exponential decay of the electric field intensity on the surface of the device, as a function of the thickness of the SiO$_2$ spacer, as calculated by RCWA. The decay length of the field intensity was $d_{RCWA} = 79.72$ nm. (b) Experimentally determined falloff of the fluorescence enhancement factor for Cy-5 as a function of the thickness of SiO$_2$ layer deposited. The decay length determined by experiment was $d_{exp} = 84.78$ nm. Both calculations and experiments were performed for an incident wavelength of $\lambda = 632.8$ nm.

Figure 4. Fluorescence intensity as a function of wavelength for semiconductor quantum dots embedded within a photonic crystal surface, where the photonic crystal resonance coincides with the emission wavelength of the quantum dots for enhancing light extraction. Compared to quantum dots on a plain surface (black curve), linear 1D, square 2D, and hexagonal 2D photonic crystal slabs provide >10x greater detection sensitivity.
ACKNOWLEDGMENT

The authors thank SRU Biosystems and the National Science Foundation for the funding that supported this work.

REFERENCES


