A simple strategy for improving fluorescent immunoassays using photonic crystals

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The most widely used substrates for fluorescent immunoassay are based on transparent materials such as glass or plastic with almost no optical functionality.
Very recently, optical nanostructures made of metal on dielectrics have been proposed to perform photon management features mediated by surface or localized plasmons. In particular, resonant ring-shaped antennas have been demonstrated to enhance the amount of overall fluorescence radiated from few emitters and provide some control on the emission direction [1]. However, such an approach based on plasmonic structures is limited by the material absorption, especially in the visible region of the spectrum.
As an alternative strategy, photonic crystals have been used instead [2]. In that case, it is possible to resonantly couple a laser radiation to specific leaky modes (e.g. in the Kretschmann configuration or via grating coupling) in such a way that an increase of fluorescence excitation is produced because of a near-field enhancement. Conversely, depending on the orientation of the dipole momentum of emitters, a significant portion of emitted light can couple to leaky modes of the photonic crystals, and being detected under specific collection angles.

In this contribution, we propose an hybrid approach for simply improving the fluorescence radiation that can be excited and extracted on a glass-based substrate for immunoassay sensing. We take inspiration from plasmonics and photonic crystals and propose the use of surface modes sustained on planar multilayers made of different dielectric materials. Although the multilayer is an example of one-dimensional photonic crystals (1DPC), the modes involved in the enhancement mechanism are very similar to surface plasmons, with some additional advantages such as: (i) the possibility of having either TE or TM polarized modes, (ii) a higher Q factor, (iii) a higher near-field intensity on the 1DPC surface. This modes, called Bloch Surface Waves (BSW) can also act as “drain” channels for the radiation emitted by organic dyes placed on the 1DPC surface [3]. This phenomenon, recalling the well known Surface Plasmon Coupled Emission (SPCE) is gaining popularity in the biosensing domain [4].

As a practical application of this mechanism, ring-shaped gratings are fabricated on the 1DPC in such a way that they can be used for coupling an external radiation to focused BSW (Fig.1) and to diffract the BSW-coupled emission normally to the 1DPC surface. When specific receptors (antibodies) are spotted corresponding to different ring antennas arranged as an array, it is possible to detect an enhanced fluorescence by using a simple wide-field fluorescence imaging system, like in conventional immunoassay for antigene detection (Fig.2). We will show the robustness of this approach, requiring almost no alignment of the laser source and very low numerical aperture for collection.

References
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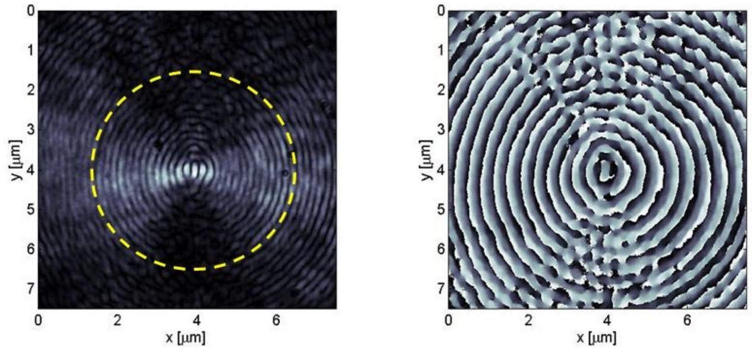

Figure 1. Interferometric leakage radiation images of the amplitude (left) and phase (right) of a surface wave focused by a circular grating on a dielectric 1DPC at λ=532 nm. Illumination is provided by a low-divergence laser beam normally impinging on the structure from the bottom. Directly transmitted light is Fourier-filtered by a circular amplitude mask.

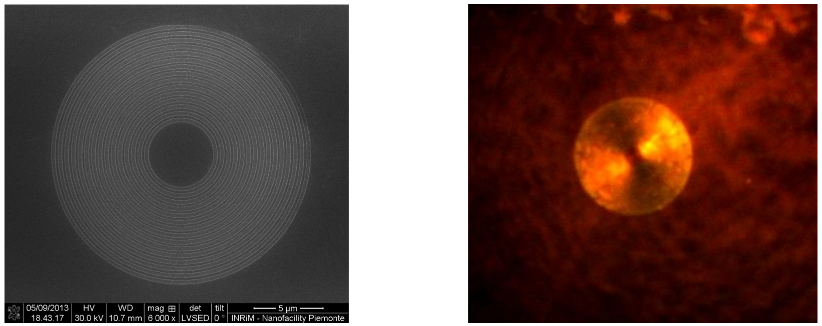

Figure 2. (left) SEM picture of the circular grating on the 1DPC; (right) fluorescence image of the ring grating on 1DPC illuminated with a collimated laser from the bottom. A R6G dye is spun homogeneously on the surface in such a way that the fluorescence enhancement can be appreciated on the ring structure as compared to the background emission.