Lossy mode resonance enabling ultra-low detection limit for fibre-optic biosensors (INVITED)

F. Chiavaioli[1]*, A. Giannetti[1], S. Tombelli[1], C. Trono[1], I. Del Villar[2], I. R. Matias[2], P. Zubiate[3], C. R. Zamarreño[3], F. J. Arregui[3], F. Baldini[1]

[1] Institute of Applied Physics "Nello Carrara," National Research Council, Via Madonna del Piano 10, 50019 Sesto Fiorentino, Firenze, Italy
[2] Institute of Smart Cities, Public University of Navarra, 31006 Pamplona, Spain
[3] Electrical and Electronic Engineering Department, Public University of Navarra, 31006 Pamplona, Spain

* f.chiavaioli@ifac.cnr.it

Abstract. The combination of optical fibre-based biosensors with nanotechnologies is providing the opportunity for the development of *in situ*, portable, lightweight, versatile and high-sensitivity optical sensing platforms. We report on the generation of lossy mode resonances (LMRs) by means of the deposition of nm-thick SnO_2 film on optical fibres. This allows measuring precisely and accurately the changes in refractive index of the fibre-surrounding medium with very high sensitivity compared to other optical technology platforms, such as long period grating or surface plasmon resonance. This approach, mixed with the use of specialty fiber structures such as D-shaped fibres, allows improving the light-matter interaction in strong way. Different imaging systems, i.e. SEM and TEM along with X-EDS tool, have been used to study the optical features of the fiber coating. The shift of the LMR has been monitored in real-time thanks to conventional wavelength interrogation system and ad-hoc developed microfluidics. A big leap in performance has been attained by detecting femtomolar concentrations in human serum. The biosensor reusability has been also tested by using a solution of sodium dodecyl sulphate.

Keywords: Lossy mode resonance, in-fiber optical sensing platform, nanofilm deposition, femtomolar concentration, reusability

1 Introduction

Compact, portable, lightweight and high sensitivity devices are more and more demanded by industrial companies as well as novel sensing concepts are continuously studied in fundamental research. Great advantages are offered from fibre-optic devices over other optical technology platforms thanks to the peculiarities of optical fibres [1,2]. Moreover, the opportunity of depositing nanometer films on optical fibres with a high degree of accuracy, precision and reproducibility has allowed the broadening of the application domains of this technology [3,4].

One of the most widespread phenomenon related to metallic thin films for sensing is surface plasmon resonance (SPR) [5]. More recently, starting from the studies on semiconductor waveguides [6], the concept of guided mode resonance was also applied to fibre-optic, under the name of lossy mode resonance (LMR) [7]. LMR occurs when the real part of the thin film permittivity is positive and greater in magnitude than both its own imaginary part and the permittivity of the material surrounding the thin film [8]. Therefore, metallic oxides and polymers are used to generate LMRs [9], instead of the metallic materials typically used to generate SPRs [10]. Differently from SPR, both TE- and TM-polarized light can excite LMR. Another important characteristic is the possibility of tuning the spectral position of the LMR by adjusting the thin film thickness [7].

LMR-based sensors have been recently implemented in D-shaped single-mode fibres instead of using multi-mode fibres, where LMRs are broader and more difficult to analyse [11]. Due to the asymmetric shape of the transversal section of the D-shaped fibre, it is possible to excite more than a single LMR, thus it allows tracking the wavelength shifts of the first LMR, the most sensitive LMR, at wavelengths in the infrared region, where the sensitivity is also improved if compared to the visible region [12].

Biosensing with optical fibres should gain a great push from this technology, since the requirement of very low limit of detection (LOD) is high in many applications. This causes in turn a change.

In this paper, we present the performance of SnO_2-coated D-shaped fibre-optic LMR biosensor by developing an IgG/anti-IgG immunological assay. The sensing principle is quite simple: when a target analyte interacts with the fibre surface firstly coated with an nm-thick film and then suitably functionalised, this induces a change in the spectral position of the LMR that can be measured accurately and precisely by means of a conventional wavelength interrogation system and an ad-hoc integrated microfluidics [12]. The results detail that it is possible to achieve femtomolar concentrations of the analyte.

2 Materials and Methods

The fibre-optic used here was a D-shaped single-mode fibre (SMF) purchased from Phoenix Photonics Ltd. (Birchington, UK). This fibre consisted of a standard SMF (SMF-28, Corning®) with a side-polished length of 17 mm (Fig. 1). Afterwards, the polished surface of the fibre, hereafter called sensitive region, was coated with a thin film of SnO_2 with a 99.99% of purity and purchased from ZhongNuo Advanced Material Technology Co. The D-shaped SMF was carefully handled and placed on an ad-hoc developed substrate in a DC sputter machine (ND-SCS200, Nadetech S.L.). The fabrication parameters were: 9×10^{-2} mbar argon partial pressure and 90 mA current intensity. The experimental setup is sketched in Fig. 1. It consisted of a broadband multi-LED light source (FIBRELABS, Inc., SLD-1310/1430/1550/1690), an optical spectrum analyser (OSA, Anritsu MS9030A-MS9701B), an in-line polarizer and a polarization controller, which allowed exciting the selected TE or TM polarized travelling light.

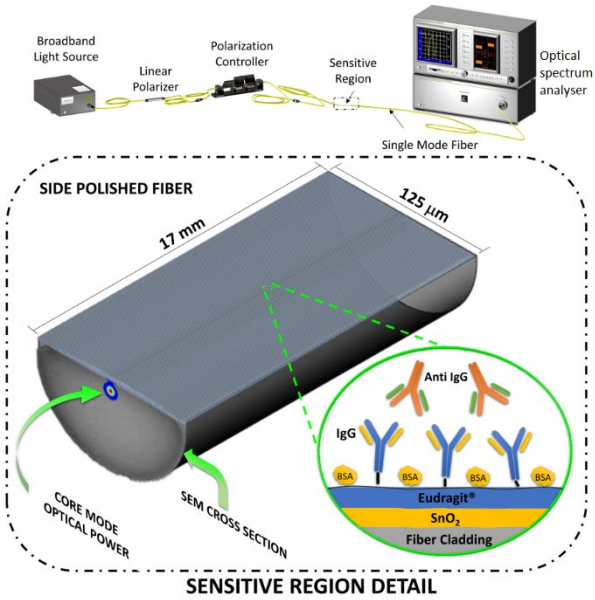

Fig. 1. Experimental wavelength interrogation system consisting of a broadband light source, a linear polarizer, a polarization controller, an optical spectrum analyser and the sensitive region (D-shaped fibre biosensor). The sensitive region along the D-shaped fibre, the device structure and immunosensing protocol are also detailed.

The functionalisation of the sensitive region was achieved by the deposition of a co-polymer (Eudragit L100, Evonik Degussa GmbH) that provided free functionalities, necessary for the antibody immobilization. The fibre was immersed in 2 mM (0.04% w/v) Eudragit L100 in ethanol for 1 minute and then let the solvent evaporate in the air for about 15 minutes. Afterwards, the fibre was inserted inside a thermo-stabilised microfluidics. Ref. [13] detailed all the steps performed for the preparation of the biological sensing layer, with the assay completed with the injection of the specific analyte at increasing concentrations from 1 ng L^{-1} up to 10 mg L^{-1}. The biosensor specificity was evaluated by spiking the analyte in CRP-free human serum.
A TEM (CM 12 PHILIPS) equipped with an OLYMPUS Megaview G2 camera and a field emission SEM (FESEM, UltraPlus Carl Zeiss Inc.) were used to measure the film thickness. X-ray energy dispersive spectroscopy (X-EDS) tool of FESEM was also used to feature the chemical elements present in the deposited thin film.

3 Results and Discussion

The features of SnO_2 thin film were firstly studied. A high magnification FESEM image of the functionalised sensor is detailed in Fig. 2a, whereas a SEM image and the corresponding X-EDS spectrum taken in correspondence of the thin film are detailed in Figs. 2b and 2c, respectively. Figs. 2d and 2e account for the TEM images of another similar sensor coated with SnO_2 film and SnO_2+Eudragit layers,

respectively. An averaged film thickness of (160±5) nm was obtained for the first case and it increased up to (220±5) nm after the deposition of the polymer, deducing a thickness of the polymer of roughly 60 nm.

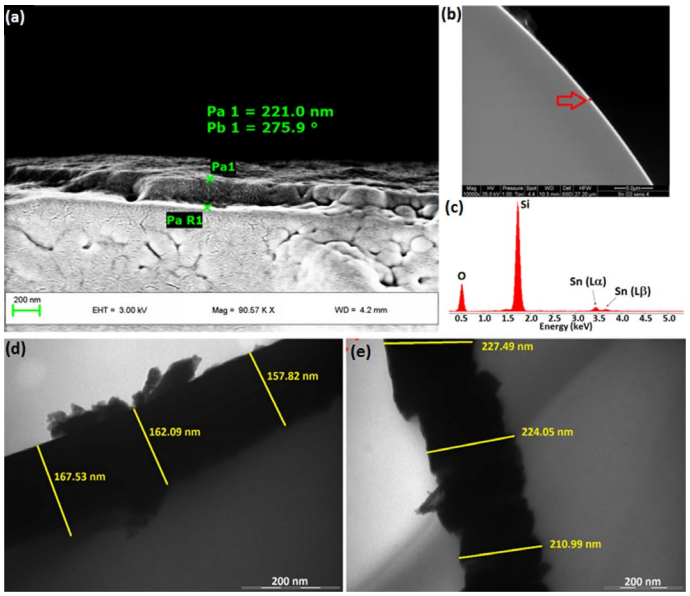

Fig. 2. (a) FESEM image of the cross-section of a D-shaped fiber coated with both SnO_2 thin film and Eudragit layer. (b) SEM image under less magnification taken for X-EDS microanalysis. (c) X-EDS spectrum of the elements present in the thin film region detailing the spectral energy lines of interest. TEM images of the cross-section of a D-shaped fiber coated with SnO_2 film (d) and with Eudragit layer too (e).

The implementation of the IgG/anti-IgG immunological assay was then carried out to monitor in real-time the binding interactions using SnO_2-coated LMR fibre biosensors. The biochemical steps of the immunoassay were followed by tracking the shift of the LMR wavelength. A washing step in PBS was mandatory to measure the LMR shift due to the effective receptor-analyte bond. Figure 3 details the calibration curve showing the LMR shift as a function of the analyte concentration. The respective standard deviation of each experimental point (15 subsequent acquisitions under the same experimental conditions) is also detailed, together with the sigmoidal fit by using the Hill equation (blue curve), formally equivalent to the Langmuir isotherm, a well-accepted mathematical model used to quantify the interaction level between ligand binding sites.

By means of the calibration curve, it was possible to obtain the biosensor LOD. Considering the blank signal plus 3σ of the blank (0.18 nm), a LOD of 0.15 ng L^{-1} (1 fM) was obtained; if 3σ of the maximum standard deviation obtained from all the experimental points (0.27 nm) is considered, a different LOD of 0.6 ng L^{-1} (4 fM) was attained. Moreover, the performances were compared with other highest-performance fibre-based biosensors from the literature. Biosensors based on long period fibre gratings [13,14] claimed a LOD of the order of μg L^{-1} (tens of pM) or slightly less.

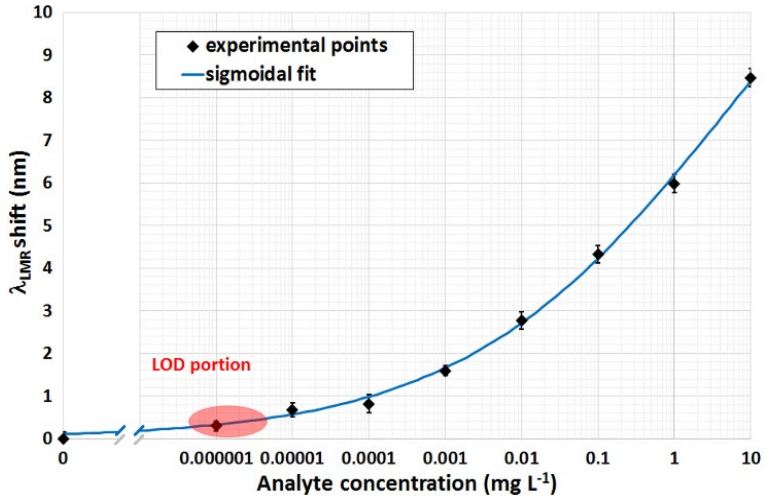

Fig. 3. Calibration curve of the SnO_2-coated LMR-based D-shaped fibre biosensor, together with the sigmoidal fit of the experimental points and their respective standard deviation (black error bars).

4 Conclusion

We proposed an optically absorbing material, i.e. SnO_2, that was deposited on D-shaped single-mode fibres for generating lossy mode resonances, which are a recently-explored physical phenomenon that allowed the development of label-free biosensors able to obtain high sensitivity and resolution in measuring refractive index changes of the fibre-surrounding medium.

TEM and SEM imaging allowed tuning the LMRs in the selected wavelength range and, hence, using a standard experimental wavelength interrogation system in NIR. In addition, the proposed biosensor was integrated into a thermo-stabilised microfluidics, thus addressing all the typical requirements that a real biosensor must possess towards the development of an *in situ*, portable, lightweight and high-sensitivity optical device for biochemical and biomedical applications.

The results have proved a big leap in performance thanks to the ability to detect analyte concentrations down to ng L^{-1} (few fM) in human serum, enhancing the LOD by three orders of magnitude when compared with other fibre-based configurations and reaching a value comparable with the best optical technology platforms, such as SPR or localised SPR.

5 Acknowledgements

This work was supported by the National Research Council of Italy (CNR) for the Short Term Mobility program 2017, by the Government of Navarra (project no.

72/2015) and by the Spanish Agencia Estatal de Investigación (AEI) and European Regional Development Fund (FEDER) (TEC2016-78047-R,TEC2016-79367-C2-2-R).

References

[1] Rajan, G., Optical Fiber Sensors: Advanced Techniques and Applications, CRC Press, Taylor & Francis Group, Chap. 1 (2015).
[2] Chiavaioli, F., Baldini, F., Tombelli, S., Trono, C., Giannetti, A., Biosensing with optical fiber gratings, Nanophotonics 6, (2017).
[3] Caucheteur, C., Guo, T., Liu, F., Guan, B.-O., Albert J., Ultrasensitive plasmonic sensing in air using optical fibre spectral combs, Nature Communications 7, (2016) 13371.
[4] Stern, L., Desiatov, B., Mazurski, N., Levy, U., Strong coupling and high-contrast all-optical modulation in atomic cladding waveguides, Nature Communications 8, (2017) 14461.
[5] Gramotnev, D. K., Bozhevolnyi, S. I., Plasmonics beyond the diffraction limit, Nature Photonics 4, (2010) 83-91.
[6] Andreev, A., Pantchev, B., Danesh, P., Zafirova, B., Karakoleva, E., Vlaikova, E., Alipieva, E., A refractometric sensor using index-sensitive mode resonance between single-mode fiber and thin film amorphous silicon waveguide, Sensors & Actuators B: Chemical 106, (2005) 484-488.
[7] Del Villar, I., Arregui, F. J., Zamarreño, C. R., Corres, J. M., Bariain, C., Goicoechea, J., Elosua, C., Hernaez, M., Rivero, P. J., Socorro, A. B., Urrutia, A., Sanchez, P., Zubiate, P., Lopez, D., De Acha, N., Ascorbe, J., Matias, J. R., Optical sensors based on lossy-mode resonances, Sensors & Actuators B: Chemical 240, (2017) 174-185.
[8] Yang, F., Sambles, J. R., Determination of the optical permittivity and thickness of absorbing films using long range modes, Jourbal of Modern Optics 44, (1997) 1155-1164.
[9] Del Villar, I., Hernaez, M., Zamarreño, C. R., Sánchez, P., Fernández-Valdivielso, C., Arregui, F. J., Matías, I. R., Design rules for lossy mode resonance based sensors, Applied Optics 51, (2012) 4298-4307.
[10] Homola, J, Surface plasmon resonance sensors for detection of chemical and biological species, Chemical Reviews 108, (2008) 462-493.
[11] Chiavaioli, F., Zubiate, P., Del Villar, I., Zamarreño, C. R., Giannetti, A., Tombelli, S., Trono, C., Arregui, F. J., Matias, J. R., Baldini, F., Femtomolar detection by nanocoated fibre label-free biosensors, ACS Sensors 3, (2018) 936-943.
[12] Chiavaioli, F., Gouveia, C., Jorge, P., Baldini, F., Towards a uniform metrological assessment of grating-based optical fiber sensors: from refractometers to biosensors, Biosensors 7, (2017) 32.
[13] Chiavaioli, F., Biswas, P., Trono, C., Jana, S., Bandyopadhyay, S., Basumallick, N., Giannetti, A., Tombelli, S., Bera, S., Mallick, A., Baldini, F., Sol–gel-based titania–silica thin film overlay for long period fiber grating-based biosensors, Analytical Chemistry 87, (2015) 12024-12031.
[14] Quero, G., Consales, M., Severino, R., Vaiano, P., Borriello, A., Diodato, L., Zuppolini, S., Giordano, M., Nettore, I. C., Mazzarella, C., Colao, A., Macchia, P. E., Santorelli, F., Cutolo, A., Cusano, A., Long period fiber grating nano-optrode for cancer biomarker detection, Biosensors and Bioelectronics 80, (2016) 590-600.