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# E. Coli detection using surface plasmon resonance

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The high sensitivity of chalcogenide surface plasmon resonance (SPR) structures with chalcogenide films is fructified to elaborate the salinity sensor for the seawater. A four layers configuration was taken into consideration, comprising a BK7 prism, a gold thin film, a chalcogenide $As_2S_3$ film and the ambient medium, which is the β-Galactosidase solution. The transfer matrix formalism was used in our study to determine the reflectances characterizing the plasmonic structure. The two polarization modes (TE and TM) were taken into consideration, in order to determine the best configuration that allows an optimal sensitivity. The structure used as optical sensor has a high sensitivity. A 0.05 % change in concentration causes an 18-arc seconds change in plasmonic resonance incident angle and it can be resolved.

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## 1. Introduction

Since Kretschmann [1] proposed the use of evanescent waves to couple surface plasmons with light, various chemical and biochemical sensors have been proposed and studied [2-5]. The coupling takes place in conditions of conservation of momentum and energy, which is reflected in a very narrow resonance curve, conducting to the development of high sensitivity optical sensors.

The Kretschmann configuration is based on a three-layer structure and the confining of the plasmon-polaritonic wave at the metal film interface with the environment. In [6] the authors included in the structure configuration a GaLaS film and a rutile prism, but this fourth layer is not a waveguide. In [7-8] we developed a structure for a four-layer configuration that also includes a thin film of semiconductor material that has a high refractive index, so that the film is a planar waveguide. The confinement of the plasmon-polaritonic wave takes place at the semiconductor film's interface with the environment. Amorphous chalcogenide compounds of the type $As_2S_3$ and $As_2Se_3$ proved to be promising for the realization of the wave guide due to the high refractive index and the well approved technology of the thin films vacuum deposition. The configuration of the electromagnetic field in certain ways of the waveguide allows to increase the sensitivity to changing the environment refractive index. It has been established that for certain thicknesses of the chalcogenide [9] film the coupling of light can also be done with prisms of crown glass like that of BK7, which has a low refractive index. The sensors based on four-layer structure can be used for water salinity determination [10].

In this paper the use of surface plasmon resonance (SPR) as biosensor to detect pathogen strains such as E. Coli bacteria was studied. It is a new approach, different from Long Period Grating Fiber Sensors [11,12], however it was preserved the idea to detect not the strain itself but a marker, - an enzyme produced by it while it is alive.

## 2. Surface plasmon resonance E. Coli detector

Surface plasmon resonance (SPR) is used as a biosensor by highlighting small changes in the refractive index of the tested liquid caused by the presence of the strain to be detected.

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As a matter of fact, the strain concentration is measured indirectly, monitoring some enzymes the strain produces. Escherichia coli produces some enzymes that can reveal the presence of the bacterium and give an indication of its concentration. These enzymes are lipopolysaccharide biochemical compounds and they are bound to the outside of the membrane of the analyzed bacterium. In our study we used β-D-galactosidase from Escherichia coli.

An Escherichia coli detector using SPR is schematically presented in Fig. 1. The tested solution (β-D-galactosidase in water) is introduced into a sealed enclosure through an intake nozzle and exhausted through an escape nozzle. The sealed enclosure ensures the contact of the tested solution with the BK7 prism hypotenuse, on which the two layers (gold and $As_2S_3$) are deposited.

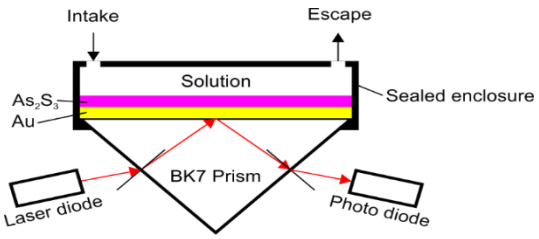


*Fig. 1. E. Coli detector schematic layout.*

The laser radiation can be generated by one of the 21 laser diodes in our library, with wavelengths ranging from 405 to 1625 nm. For this study the wavelength of 1550 nm was chosen.

Fig. 2 displays the tested solution refractive index variation with wavelengths ranging from 405 to 1625 nm, for five concentrations (0 %, 0.05 %, 0.1 %, 0.5 % and 1 %).

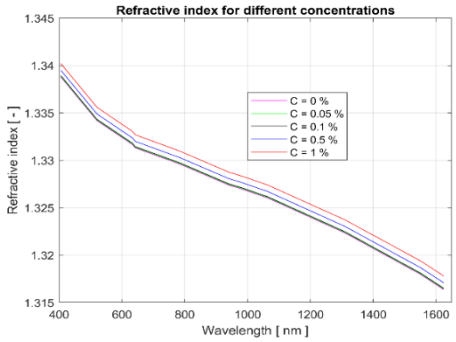


*Fig. 2. Refractive index for different solution concentrations.*

The refractive index of the solution was determined considering that the solution will have a refractive index $n_s$ which is between that of water ($n_w$) and that of β-Galactosidase ($n_g$) depending linearly of concentration C: $n_s = n_w + (n_g - n_w) \cdot C[\%]/100$.

The tested solution refractive index variation with concentration at the chosen wavelength (1064 nm) is shown in Fig. 3.

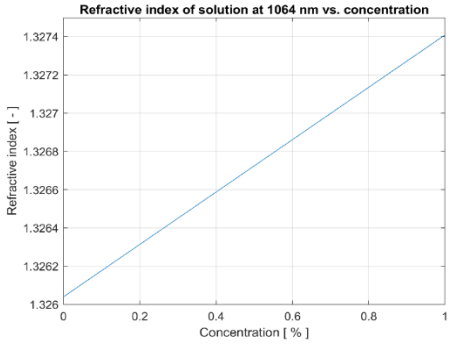


*Fig. 3. Refractive index of solution at 1064 nm vs. concentration.*

## 3. Results and discussion

A four layers configuration was taken into consideration, comprising a BK7 prism, a gold thin film, a chalcogenide $As_2S_3$ film and the ambient medium, which is the β-Galactosidase solution, having five different concentrations. The layers are described in the Table 1. Refractive indices are given for the chosen wavelength (1064 nm).

*Table 1. Layers characteristics.*

| Layer no. | Material | Refractive index | Thickness |
|---|---|---|---|
| 1 | BK 7 | 1.506635 | - |
| 2 | Au | 0.314837 + 6.628261i | 45 nm |
| 3 | $As_2S_3$ | 2.469346 | 1000 nm |
| 4 | Solution 0 % | 1.326040 | - |
| | Solution 0.05 % | 1.326108 | |
| | Solution 0.1 % | 1.326177 | |
| | Solution 0.5 % | 1.326725 | |
| | Solution 1 % | 1.327409 | |

The thickness of the gold layer was chosen to be 45 nm, a value that allows to get a low reflectance minimum. The transfer matrix formalism was used to calculate the reflectance curves characterizing the plasmonic structure [6-9,13-15]. Different thicknesses of the $As_2S_3$ layer were used to detect plasmonic resonance. Finally the plasmonic resonance was obtained for a 1000 nm thick $As_2S_3$ layer.

Five structures (described in Table 1), corresponding to the five concentrations of the solution taken into account (0, 0.05, 0.1, 0.5 and 1 %), were analyzed at the wavelength 1064 nm using a very fine mesh of incidence angles, in the range 10° - 85°, with a step of 0.005°. The two polarization modes (TE and TM) were taken into consideration, in order to determine the best configuration that allows an optimal sensitivity.

The simulation parameters are summarized in Table 2.

*Table 2. Simulation parameters.*

| Parameter | Value |
|---|---|
| Wavelength | 1064 nm |
| No. of layers | 4 |
| Starting angle | 10 ° |
| Ending angle | 85 ° |
| Angle step | 0.005 ° |

The reflectance dependence on incidence angle at different concentrations of the solution was obtained. Fig. 4 displays only the area of interest, around the plasmonic resonance angle range.

The results ($R_{min}$ – minimum reflectance, i.e. at plasmonic resonance, $\theta_{min}$ – incidence angle corresponding to the minimum and $w_{min}$ – half-measure width at minimum reflectance) are summarized in Table 3.

Surface plasmonic resonance was reached since minimum reflectances (about 1 %) and low widths at half-measure (about 0.25 °) were obtained.

*Table 3. Plasmonic resonance reflectance, incident angle and half-measure width at different concentrations.*

| Concentration [ % ] | $R_{min}$ [ % ] | $\theta_{min}$ [ ° ] | $w_{min}$ [ ° ] |
|---|---|---|---|
| 0 | 1.0255 | 61.945 | 0.250 |
| 0.05 | 1.0258 | 61.950 | 0.250 |
| 0.1 | 1.0267 | 61.955 | 0.250 |
| 0.5 | 1.0553 | 61.995 | 0.250 |
| 1 | 1.0054 | 62.040 | 0.245 |

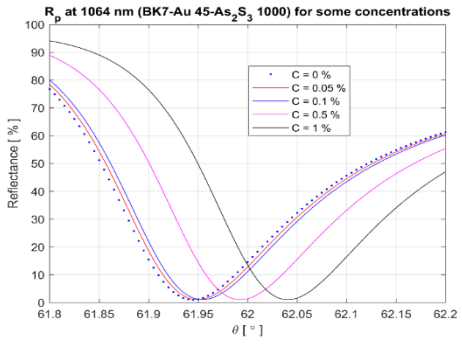


*Fig. 4. Reflectance vs. incidence angle at different concentrations.*

Fig. 5 details the dependence of plasmonic resonance incident angle with solution concentration.

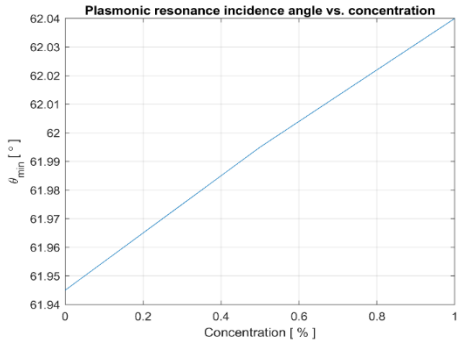


*Fig. 5. Plasmonic resonance incidence angle vs. concentration.*

The structure used as optical sensor has a high sensitivity. A 0.05 % change in concentration causes an 18-arc seconds change in plasmonic resonance incident angle and it can be resolved. A typical rotation stage with stepper motor has 409,600 microsteps per revolution, which is 3.164 arc seconds per microstep. The resulting sensitivity is then 0.0088 %.

## 4. Conclusions

The study proves that a chalcogenide surface plasmon resonance structure can be used to accurately determine the presence of the E. Coli bacterium by measuring the concentration of a marker enzyme in the tested water.

A four layers configuration was taken into consideration, comprising a BK7 prism, a 45 nm gold thin film, a 1000 nm chalcogenide $As_2S_3$ film and the tested solution (for 5 different concentrations) as ambient medium.

The plasmon resonance structure was designed in two steps. First a large range of $As_2S_3$ film thicknesses have been tried only for concentration 0 % until an optimal value (1000 nm) for this thickness was found. Thus the lowest reflectance minimum and the narrowest notch were obtained. In the second stage, the transfer matrix formalism was applied to determine the reflectance characterizing the plasmonic structure for all studied concentrations, using the $As_2S_3$ film thicknesses previously found as optimal. The large range of incidence angles (from 10° to 80°, by a very fine step – 0.005°) makes the analysis very accurate.

Plasmonic resonance reflectance, incident angle and half-measure width were determined for some concentrations, resulting a very good sensitivity. The linearity of the plasmonic resonance incident angle dependence on concentration is very good, allowing an interpolation for other concentration values. Plasmonic resonance reflectance minimum was found to be around 1 %.

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