# A self-referenced optical colorimetric sensor based on silver and gold nanoparticles for quantitative determination of hydrogen peroxide

Pedro J. Rivero [a,*], Elia Ibañez [b], Javier Goicoechea [b], Aitor Urrutia [b], Ignacio R. Matias [c], Francisco J. Arregui [b]

[a] Materials Engineering Laboratory, Department of Mechanical, Energetic and Materials Engineering, Public University of Navarre, Campus Arrosadía S/N, 31006 Pamplona, Spain
[b] Nanostructured Optical Devices Laboratory, Department of Electrical and Electronic Engineering, Public University of Navarre, Campus Arrosadía S/N, 31006 Pamplona, Spain
[c] Institute of Smart Cities, Public University of Navarre, Campus Arrosadía S/N, 31006 Pamplona, Spain

## ARTICLE INFO

## ABSTRACT

In this work, a self-referenced colorimetric sensor for the quantitative determination of hydrogen peroxide is presented. This optical sensor is based on the presence of the Localized Surface Plasmon Resonances of silver and gold nanoparticles which are capped with the same encapsulating agent of poly(diallylammonium chloride) (PDDA). These metallic nanoparticles are synthesized by a chemical reduction method of their corresponding inorganic precursors and characterized by UV–vis spectroscopy and transmission electron microscopy, respectively. A remarkable difference in sensitivity related to both LSPR absorption bands is observed as a function of variable molar concentration of the target molecule. The LSPR band of the silver nanoparticles is gradually decreased whereas the LSPR of the gold nanoparticles is practically unaltered when the hydrogen peroxide molar concentration is increased as a result of its better chemical stability. This stable absorbance LSPR band of the AuNPs is used as an optical reference and the molar concentration of the target molecule is obtained by measuring the changes of the absorbance strength maxima of both LSPR absorption bands. As a result, a very good sensitivity with a high robustness and a linear response over a wide concentration range from $1.25\,\mu M$ to $1250\,\mu M$ is obtained. This self-referenced method opens up a new perspective for the chemical detection of other reactive oxygen species or well for the determination of other type of analytes in the future.

© 2017 Elsevier B.V. All rights reserved.

## 1. Introduction

In the last decades, the rapid proliferation of the nanotechnology has led to the fabrication and use of an increasing number of nanomaterials and nanoparticles in fields as diverse as biology, catalysis, chemistry, construction, electronics, medicine, textile or optics, among others [1–8]. One of the most explored characteristics of the noble metal nanoparticles (essentially silver, gold or copper) is a phenomenon known as Localized Surface Plasmon Resonance (LSPR), the frequency at which conduction electrons collectively oscillate in response to the alternating electric field of an incident electromagnetic radiation. As a result, an intense absorption band in the visible region with a specific coloration of the resultant nanoparticles is obtained [9,10]. A wide variety of chemical routes for the synthesis of metal nanoparticles with different shapes can be found in bibliography [11–14], although the most common approach involves the chemical reduction of metal salts in the presence of an adequate encapsulating agent [15]. The encapsulating agent plays an important role for the synthesis of nanoparticles with a specific morphology because it limits the growth of the particles, directs their shape and provides colloidal stability [16].

The wavelength location of this LSPR absorption band shows a great dependence over several factors such as the resultant shape and size of the nanoparticles, the refractive index of their surrounding medium or the environmental in which the metal nanoparticles are dispersed [17–19]. In addition, other key factor is the average distance between neighboring metal nanoparticles because when the average particle distance is shortened to a distance below the diameter of the particles, an aggregation of the nanoparticles is performed. This aggregation leads to a total color change as a consequence of the LSPR coupling between nanoparticles and as a
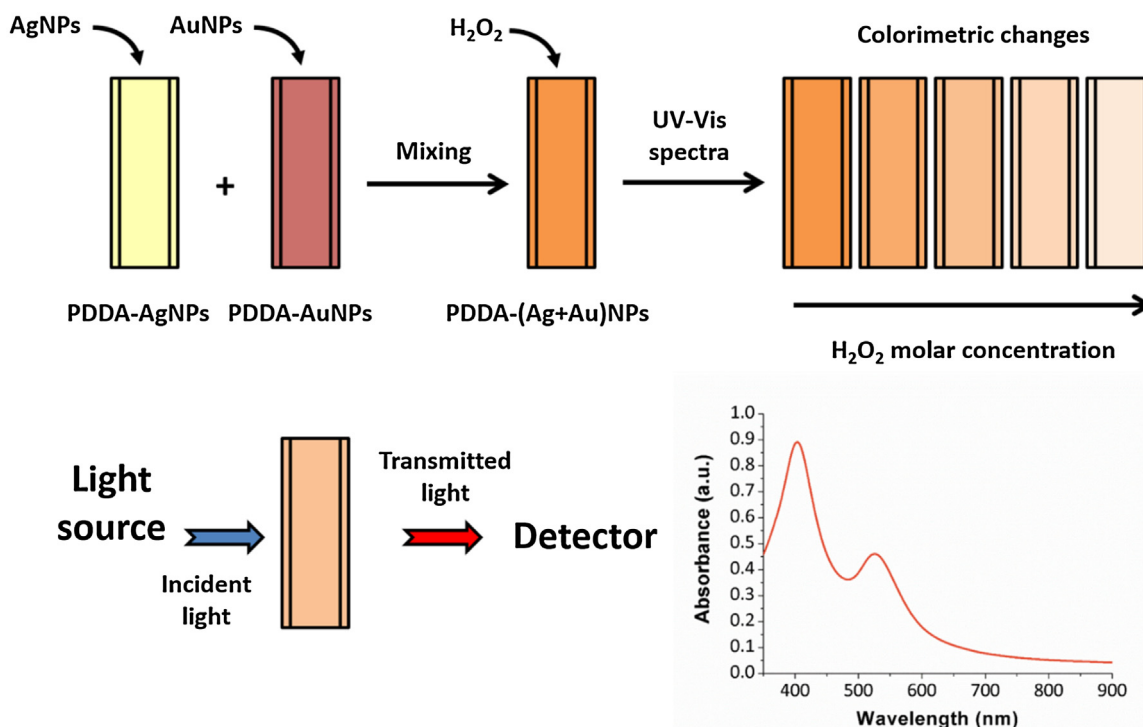
**Fig. 1.** Schematic representation of the procedure for obtaining PDDA-(Ag + Au)NPs and experimental setup used for hydrogen peroxide detection as a function of optical colorimetric changes.

result, a red-shift of the LSPR absorption band is obtained [20]. This sensing mechanism based on the aggregation of silver or gold nanoparticles is the basis for the design of a wide number of colorimetric biosensors because it is studied the change in coloration generated by the plasmon coupling between nanoparticles upon aggregation [20–25]. However, other colorimetric sensing mechanisms have used the excellent optical properties of the noble metal nanoparticles as a colorful reporter by measuring the changes in the absorbance strength for the quantitative determination of a wide variety of analytes for biological or medical purposes [26–30].

One of the analytes that are causing a great interest in the medical community is the hydrogen peroxide because it is well-known Reactive Oxygen Specie (ROS) [31–35]. An excessive accumulation of this analyte in the body can trigger some diseases because the hydrogen peroxide is the responsible of causing tissue damage and DNA fragmentation. In addition, it is the product of reactions catalyzed by a large number of oxidase enzymes, and therefore its monitorization can lead to different bio-sensing applications. Therefore, the analytical determination of $H_2O_2$ also has an important significance in other totally different fields such as industry, food or environmental analysis. According to this multidisciplinary presence, the design and development of new methodologies for the quantitative determination of $H_2O_2$ are required. Most of the analytical methods for its quantification are based on enzymes, using different techniques such as chemiluminiscence, spectrofluometry and electrochemistry [36–38]. However, these techniques show several drawbacks such as the use of high sophisticated instrumentation, costly performance, need of a total immobilization of the enzymes in order to obtain a good sensitivity, long assay time, low selectivity or a poor reproducibility. From all these backgrounds, the use of other alternative method is required for the determination of this target molecule. Among all them, LSPR optical sensors based on the incorporation of metal nanoparticles are an ideal candidate due to their high sensitivity, great selectivity, simplicity and cost effective detection. Previous works have reported the quantification of hydrogen peroxide by using LSPR optical col-

orimetric sensors [26,32,39–42] based on the optical changes of a unique LSPR absorption band related to only silver nanoparticles.

In this work, it is reported for the first time a dual reference LSPR optical colorimetric sensor based on the combined colloidal dispersions of gold and silver nanoparticles with the same encapsulating agent for the quantitative determination of hydrogen peroxide. The metallic nanoparticles have been synthesized by a chemical reduction step from their corresponding inorganic salts. From the experimental results, it is concluded that a remarkable difference in sensitivity related to both LSPR absorption bands is observed as a function of variable molar concentration of the target molecule, making possible the design of a robust self-referenced LSPR optical detection method.

As a promising result, a very good sensitivity and a linear response over a wide concentration range from $1.25\,\mu M$ to $1250\,\mu M$ of target molecule is presented. In addition, the experimental results show a lower quantification limit than previously reported enzyme-based biosensors [43–48]. The hydrogen peroxide concentration range of the biosensor presented in this work is suitable for some biomedical applications such as oxidative stress estimation, measuring the hydrogen peroxide in human urine [31], as a simplified test for bio-monitoring the glucose in blood or human serum [26,33,35,40], or even for heavy metal ion detection such as mercury due to the catalytic reactivity of AgNPs [34]. Finally, the resultant dual LSPR optical sensor opens up a new perspective for the design and development of biosensors for monitoring other reactive oxygen species or well other analytes in the future.

## 2. Experimental section

### 2.1. Reagents and materials

Poly(diallyldimethylammoniumchloride) (PDDA) (Mw. 15000), silver nitrate (>99% titration) solution in water 0.1 N, gold (III) chloride trihydrate ($HAuCl_4\cdot3H_2O$), borane dimethylamine complex (DMAB) and hydrogen peroxide ($H_2O_2$) were purchased from