

## Abstract

# Superoxide Dismutase Determination on Silver Nanostructured Substrates through Surface-Enhanced Photoluminescence <sup>†</sup>

Anastasia Kanioura <sup>1,\*</sup>, Georgia Geka <sup>1</sup>, Ioannis Kochylas <sup>2</sup>, Vlassis Likodimos <sup>2</sup>, Spiros Gardelis <sup>2</sup>,  
Anastasios Dimitriou <sup>3</sup>, Nikolaos Papanikolaou <sup>3</sup>, Sotirios Kakabakos <sup>1</sup> and Panagiota Petrou <sup>1</sup>

<sup>1</sup> Immunoassays/Immunosensors Lab, Institute of Nuclear & Radiological Sciences & Technology, Energy & Safety, NCSR “Demokritos”, 15341 Aghia Paraskevi, Greece; g.geka@rrp.demokritos.gr (G.G.); skakab@rrp.demokritos.gr (S.K.); ypetrou@rrp.demokritos.gr (P.P.)

<sup>2</sup> Section of Condensed Matter Physics, Department of Physics, National and Kapodistrian University of Athens, University Campus, 15784 Athens, Greece; ikochyla@phys.uoa.gr (I.K.); vlikodimos@phys.uoa.gr (V.L.); sgardelis@phys.uoa.gr (S.G.)

<sup>3</sup> Institute of Nanoscience & Nanotechnology, NCSR “Demokritos”, 15341 Aghia Paraskevi, Greece; a.dimitriou@inn.demokritos.gr (A.D.); n.papanikolaou@inn.demokritos.gr (N.P.)

\* Correspondence: nkanioura@ipta.demokritos.gr

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**Abstract:** Oxidative stress is defined by an imbalance between the generation of reactive oxygen species and the biological system’s ability to neutralize them. This condition is commonly linked to various pathological conditions [1]. Superoxide dismutase (SOD) is a widely used enzyme to assess oxidative stress, and various techniques have been developed for its detection in biological samples such as blood, urine, and saliva [2]. Surface-enhanced photoluminescence (PL) is a particularly sensitive method, offering minimal interference from the sample matrix [3]. In this work, silver nanostructured surfaces were implemented as substrates for the immunochemical determination of SOD in synthetic saliva through PL. The substrates were prepared using a single-step metal-assisted chemical etching method (MACE), resulting in the formation of silicon nanowires decorated with silver dendrites of approximately 1.5  $\mu\text{m}$  in height [4]. For SOD detection, a three-step competitive immunoassay configuration was followed. Briefly, SOD was immobilized onto the substrates and then the functionalized substrates were incubated with mixtures of SOD with anti-SOD primary antibody, prepared either in assay buffer or synthetic saliva. Then, a solution of biotinylated anti-species specific antibody was added, followed by a reaction with streptavidin labelled with the fluorescent dye Rhodamine Red-X, and the signal was determined through an in-house developed optical set-up. The developed method presents similar or slightly lower sensitivity (detection limit 0.05  $\mu\text{g/mL}$ ) compared to the literature; however, it does not require labor-intensive sample pretreatment steps [5,6]. The aforementioned findings demonstrate the capability of the developed method to detect superoxide dismutase in natural saliva, in order to evaluate the oxidative stress status of an organism.

**Keywords:** superoxide dismutase; photoluminescence; oxidative stress; saliva



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