Al-doped zinc oxide (AZO) has a great potential on biosensing devices due to its low-cost, high electrical conductivity, visible transparency and better stability than other oxides such as indium tin oxide (ITO) [1]. It can be functionalized with thiol Self-Assembled Monolayers (SAMs) in order to fabricate new sensor architectures that integrate optical, electrical and electrochemical detection methods [2]. Glutathione is the most abundant non-protein thiol tripeptide and is present in almost all living organisms, predominantly in eukaryotic cells. It has many important roles, such as detoxification and protection against oxidative stress due to the reactive oxygen species [3].

In this work, AZO films and AZO/Au bilayer structures were deposited by rf-magnetron sputtering on glass and quartz substrates. Anti-Glutathione antibodies are covalently immobilized on the AZO surface modified by 3,3′-dithiodipropionic acid di(N-hydroxysuccinimide ester) (DTSP) SAM. The resulting structures were studied by spectroscopic ellipsometry in external reflection and total internal reflection modes. These techniques allowed us to estimate the SAM and antibody thicknesses and to carry out studies on the kinetics of the SAM formation. There was a significant change in the ellipsometric parameters, psi and delta, when the antibody is further bound on the thiol monolayer (Figure 1a). To prove the presence of the antibody, gold nanoparticles capped with glutathione were deposited on the antibody layer. Scanning electron microscopy (SEM) was used to analyze the morphology of the resulting surfaces confirming 1) the presence of the antibodies through their darker and gelatinous structure, and 2) the presence of gold nanoparticles, as white dots on the antibody layer (Figures 1b and 2).

The developed biosensing platforms will be employed to perform real-time measurements of glutathione using total internal reflection ellipsometry in several types of samples. For this purpose, a flow cell has been constructed to enable surface functionalization during ellipsometric characterization. Complex samples such as the bloodstream or plant oils will be analyzed in the future to assess the potential interest for sensor commercialization.

Figure 1. a) Ellipsometric spectrum. Psi and Delta parameters are represented for AZO surface (solid lines) and for anti-Glutathione-DTSP-AZO surface (dash lines). b) Typical SEM image of immobilized anti-Glutathione. Darker and lighter areas represent antibody and AZO layers, respectively. The schematic of the monolayer formed is shown beside it (glass substrate, AZO film, DTSP monolayer and antibody layer).

Figure 2. Typical SEM image of immobilized anti-Glutathione with gold nanoparticles. A gelatinous structure can be observed corresponding to the antibody layer and white dots are gold nanoparticles. The schematic of the monolayer formed is shown beside it.