THERMAL-NANO-INMMUNOASSAY (THERMOELISA) FOR THE DETECTION OF CANCER BIOMARKERS

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Gold nanoprisms (AuNPrs) are anisotropic nanoparticles that exhibit localized surface plasmon resonance (LSRP) in the near infrared (NIR) region. This optical property allows low concentrations of AuNPRs to convert light energy into quantifiable heat (photothermal effect) and makes their application interesting in the development of biological assays. AuNPrs linked to a biological receptor can act as physical-chemical transducers, interpreting the biological recognition with the analyte as a quantifiable thermal signal, that can be detected with an IR camera.

Nowadays, gastric cancer is one of the leading causes of cancer-related death worldwide. It has a poor prognosis, with a low 5-year survival and a median overall survival of less than one year in the case of advanced stages. The identification of unique patterns and specific biomarkers is essential to develop accurate diagnosis tools and effective treatments. Until now, the number of effective biomarkers applied in the clinic for the diagnosis of gastric cancer is very low.

The objective of this work is to develop a simple, economical and accessible diagnosis platform with high sensitivity, based on AuNPrs thermal detection and applicable to different tumor markers to improve current methods.

To evaluate the possibilities of the thermo-sensing technology proposed in this clinical application, a new sandwich ELISA immunoassay has been developed using the carcinoembryonic antigen (CEA) as a model analyte.

For this goal, the union of the detection antibody (Ab_det) with AuNPrs have been optimized by means of the oriented binding AuNPrs@streptavidin – biotin@Ab_det. This strategy provides a versatile bioconjugate that can be subsequently linked to any biotinylated antibody which allows extending the methodology to other cancer biomarkers. Finally, in the presence of the analyte to be detected, a "sandwich" of biomolecule-nanoparticles is formed and finally, the irradiation with a NIR laser source allows thermal transduction of the biological signal (ThermoELISA immunoassay). (Figure 1)

In this way, the final ThermoELISA developed greatly increases the sensitivity with respect to the classic colorimetric ELISA and decreases the limits of detection (0.91 ng/mL) and quantification (3.03 ng/mL) with a low RSD (13%) and a less time-consuming. This methodology has been applied to the determination of others gastric tumor markers from blood, carbohydrate antigen 19-9 (CA 19-9) and vascular Endothelial Growth Factor (VEGF), obtaining better results also to the classic colorimetric methods. Currently the methodology is being evaluated and applied to real samples.

These results highlight the transduction capacity of these nanomaterials in biological assays, as well as their possibilities in the development of biosensors.

However, this methodology requires thermal sensors (infrared camera or thermo-resistances) to quantify the signal, which makes its practical application in medical centers more complex. For this reason, we are currently working in the combination of this thermal properties, with fluorescent detection to be able to transfer heat changes to fluorescent signal. The union of both transductions aims to simplify the detection, further increase the sensitivity of the method and further reduce the limits of detection.

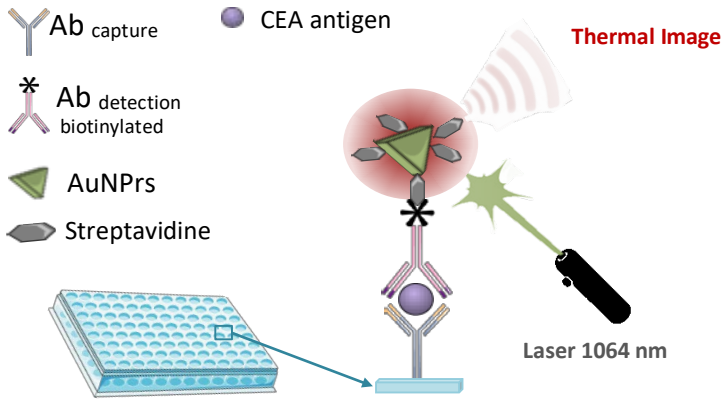

Figure 1: Schematic ThermoELISA sandwich