



Single nanoparticle ICP-MS for characterization of gold nanoparticles. A potential tool for bioassay

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1. Introduction

Nanotechnology stands as one of the fastest-growing fields in both science and technology. The synthesis of metal nanoparticles constitutes an active research area within nanotechnology, demonstrating exponential progress in their biomedical applications, particularly in imaging, diagnostics, drug delivery, and therapeutics [1, 2]. Gold nanoparticles (AuNPs) hold immense potential due to their unique optical, electrical, and catalytic properties. These properties have led to their widespread applications in medicine, electronics, and modern material manufacturing technologies [3,4]. Notably, the utilization of AuNPs as drug delivery systems is a rapidly expanding field due to their inherent properties making them highly promising drug carriers. Additionally, controlled fabrication techniques have established the creation of various sized particles (1-150 nm) with limited size dispersity. Furthermore, ligand exchange reactions have enabled the fabrication of multifunctional monolayers. This structural diversity enables particle surfaces to contain multiple targeting agents and/or chemotherapeutics [5]. Adding to that, AuNPs have been highly used as agents in biomedical detection and contrasting image due to its properties that match: biocompatibility, bioconjugation and optical properties. Their small size, high surface area and stability at high temperatures make them perfect tool in medical diagnostics, photodynamic therapy as well as in the active transport of drugs, especially for cancer treatment [6], minimizing the damage cell during optical staining or biomarker tagging even as during the delivery of biomolecules/drugs to the target cell/tissue/organ [7]. Gold nanoparticles have been employed in radiotherapy, in two modes: to increase local dose deposition in tissue during radiotherapy or as a local emitter of gamma and beta rays. The radioactive properties of gold include: ^{198}Au ($\beta_{\text{max}} = 0.96$ MeV; $t_{1/2} = 2.7$ days) and ^{199}Au ($\beta_{\text{max}} = 0.46$ MeV; $t_{1/2} = 3.14$ days), making it a strong candidate to therapeutic radio applications as well [8]. Furthermore, both isotopes have gamma emission that can be used in pharmacokinetic and dosimetric studies. Theranostics is defined, therefore, as a combination of therapeutic and diagnostic methods to describe a new platform in both treatment and diagnosis. Hybrid AuNPs have numerous applications in theranostic-based nanomedicine. The active surface and unique characteristics of AuNPs give them extensive applications in cancer and so infectious disease studies. Many works in theranostics and synthesis and characterization of AuNPs have been done in Institute (Instituto de Pesquisas Energéticas e Nucleares - IPEN-SP) [2, 3, 9,10]. So, accurate size measurement is paramount for the reliability of obtained results for nanomaterial characterization as it is one of the nanomaterial characteristics which influences their biodistribution after entry in the body. The evaluation of drug nanodelivery systems must be considerably executed to ensure their optimal performance. For this aim, there is a growing need to develop analytical strategies that permit to control the different parts of the process, from the characterization of the nanodelivery system to its behavior in biological systems like cells or tissues [11]. One technique that is proving invaluable for detecting and sizing metallic nanoparticles is the single particle inductively coupled plasma mass spectrometry (sp-ICP-MS) [12]. Its combination of elemental specificity, sizing resolution, and unmatched sensitivity makes it extremely applicable for the characterization of nanoparticles containing elements such as Au, Ag, Ti (TiO_2), and Si (SiO_2), to name a few, which have been integrated into larger products such as consumer goods, foods, pharmaceuticals, and personal care products. sp-ICP-MS is showing a great deal of promise in several application areas, including the determination of gold nanoparticle concentrations in complex samples [13]. This technique offers the unique ability to differentiate between the analyte in solution and its nanoparticle form, eliminating the need for prior separation techniques. This simplifies nanoparticle analysis by removing complex sample preparation steps [14]. Consequently, sp-ICP-MS can simultaneously provide information on nanoparticle size and size distribution, as well as the dissolved concentration of the analyte and the particle concentration (parts/mL).

A new software program for nanoparticle characterization was recently installed on an inductively coupled plasma mass spectrometer (ICP-MS) located in the 'LCQI' mass spectrometry laboratory at the Center for Lasers and Applications (CELAP) at IPEN. This software, single particle ICP-MS (sp-ICP-MS), allows for the analysis of individual nanoparticles. Our Institute holds a strong interest in researching the synthesis of gold nanoparticles (AuNPs) for various applications, including cancer treatment and cosmetology. Within this context, the present paper propose the characterization size of AuNPs by sp-ICP-MS. The ultimate goal of our research is to develop stable and well-characterized nanoparticles.

2. Methodology

The single particle inductively coupled plasma mass spectrometry was carried out using the NexION 300D (Perkin Elmer, USA). The NexION instrument is an inductively coupled plasma mass spectrometer that performs elemental analysis of varied samples. The system consists of a benchtop ICP-MS instrument, roughing pump, recirculator, cooler, and data acquisition and analysis software. Instruments equipped with Universal Cell Technology™ (UCT™) functionality may be operated in Dynamic Reaction Cell™ (DRC™) mode for ultimately low detection limits, or in easy-to-use Kinetic Energy Discrimination (KED) mode for rapid analysis. The Syngistix™ Nano Application Module is the software that combines real-time single particle acquisition with fast data processing. Solutions of AuNP were made at mass concentrations in the ng mL⁻¹ range. For calibration curve of size for sp-ICP-MS results, the gold nanospheres PEG-COOH 30, 50 and 100 nm size standard (NanoComposix) were used. A Spex plasma standard was used for calibration of dissolved gold. Dwell Time was 50 μs, Scan Time was 100s, Sample Flow Rate was 0.342 mL min⁻¹ and Transport Efficiency was 6.42.

Dynamic light scattering was carried out using Litesizer 500 Series Instrument (Anton Paar GmbH, Austria), which was equipped with a 658 nm laser and dynamic light-scattering (PCS) at 90° for particle sizing. The particle size (multimodal size distribution) was determined by measuring the angles in which an incident light beam is scattered as a function of Brownian motion of the colloidal gold particles.

NanoComposix carried transmission electron microscopy (TEM) out using JEOL 1010 Transmission Electron Microscope.

For the quality control of analyses by sp-ICP-MS, standard material 80 nm silver shelled gold nanospheres NanoXact (Au@AgNP) sold by nanoComposix was used. The certificate provided the diameter total (TEM) 79 ± 9 nm, core diameter (Au) 51 ± 6 nm and shell thickness (Ag) 14 nm. The hydrodynamic diameter (DLS) reported was 85 nm.

3. Results and Discussion

For DLS technique, validated by Gerald *et al.*, [15] the hydrodynamic diameter determinate was 84.91 ± 3.74 nm in agreement to nanoComposix report. The silver shelled gold nanospheres NanoXact (Au@AgNP) were classified monodisperse fig. 1a. The success of sp-ICP-MS analysis for determined Au@AgNP size were shown in fig. 1b. The average diameter provided by sp-ICP-MS technique was 52 ± 10 nm. The certificate provided core diameter (Au) 51 ± 6 nm and thus the technique sp-ICP-MS agrees with the reported core diameter. The sp-ICP-MS technique emerges, therefore, to complement the characterization of the nanoparticles produced.

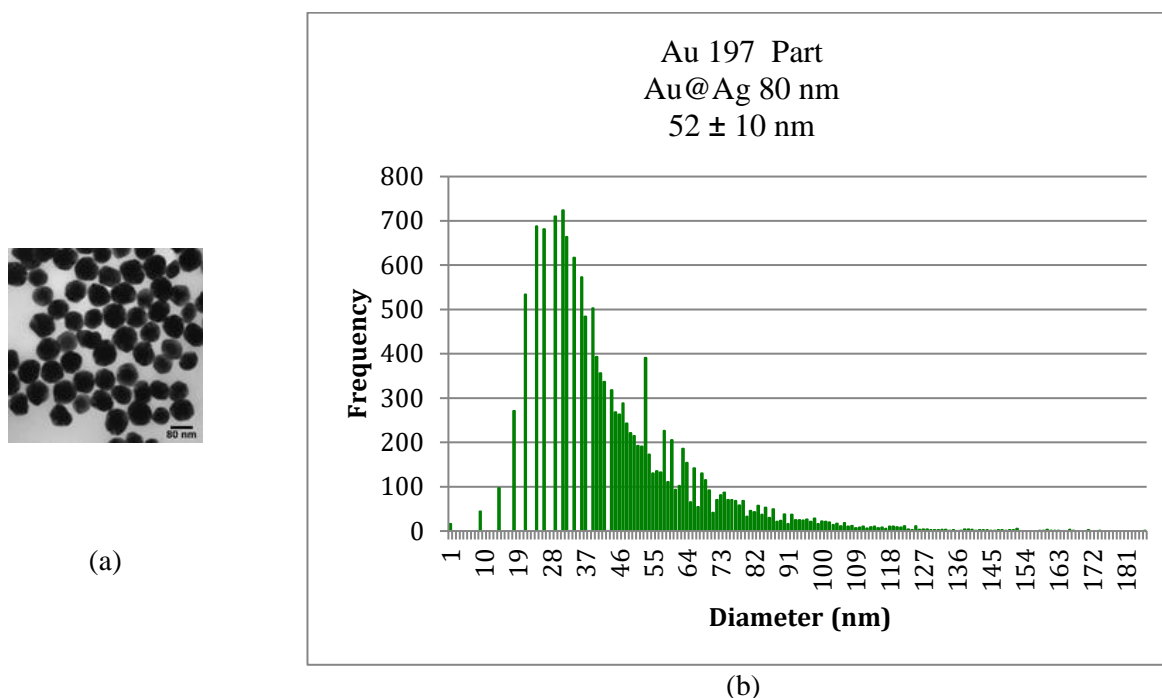


Figure 1: (a) TEM micrograph of Au@AgNP by NanoComposix. (b) Size distribution of Au@AgNP nanoComposix by ICP-MS in a single particle mode.

4. Conclusions

This paper reports our results on the gold nanoparticles characterization operating in single particle mode, sp-ICP-MS, provided a potential tool for bioassay. The technique recently implemented in our laboratory opens up the possibility of develop analytical methodology that permit to control the different parts of the process of development of new nanomaterials, from the characterization of the nanodelivery system to its behavior in biological systems like cells or tissues.

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