

AF4-ICP-MS as a powerful tool for the separation of gold nanorods and nanospheres

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Abstract

An analytical methodology based on asymmetric flow field flow fractionation (AF4) hyphenated to inductively coupled plasma mass spectrometry (ICP-MS) has been developed to separate gold nanorods (AuNRs) and nanospheres (AuNSs). This separation is of special interest because the latter are the main impurity in the synthesis of AuNRs by seeding growth procedures, and quality control tools are highly demanded. AF4-ICP-MS separation was performed using a regenerated cellulose membrane (molecular weight cut-off, MWCO, of 10 kDa), a 350 μm thick spacer and a phosphate buffer (1 mM NaH_2PO_4 , 1 mM Na_2HPO_4 at pH 7) with 0.01 % SDS as carrier. Several experimental parameters, such as focusing conditions and elution cross flow, and the injection/focusing time were optimized. Working under the optimum separation conditions, an adequate resolution was achieved between the void peak, commercially available AuNSs and AuNRs. The optimized method was successfully applied to the analysis of the products obtained in the synthesis procedure for AuNRs of 4.6 aspect ratio. Absorption molecular spectroscopy and transmission electron microscopy (TEM) have been used as complementary techniques.

Keywords

Gold nanoparticles; Nanospheres; Nanorods; Asymmetric flow field flow fractionation; Inductively coupled plasma mass spectrometry.

1. Introduction

Nowadays, gold nanoparticles (AuNPs) attract tremendous interest due to their unique properties (optical, electrochemical, magnetical, and catalytic), chemical stability and easy synthesis and functionalization¹. Size, shape, composition and structure are the main characteristics that determine these properties. For instance, it is known that gold nanospheres (AuNSs) in aqueous solution exhibit an absorption peak from 500 to 550 nm¹, while octahedral- and triangle-shaped AuNPs show bands between 580 and 600 nm², and gold nanorods (AuNRs) present two bands in the visible and near infrared regions^{3,4}. These properties lead to promising applications of AuNPs in different fields, such as sensing, imaging and biomedicine^{1,3,5}.

Among the AuNPs, the AuNRs are of special interest because they play a key role in very important biomedical applications, and both top-down and bottom-up synthesis procedures have been proposed^{6,7}. Typical bottom-up methods include wet-chemical, electrochemical, sonochemical, solvothermal, microwave-assisted, and photochemical reduction techniques⁴. A hard or soft template, which serves to confine the growth along one direction, is required in these procedures and the most commonly used soft template is seeding-mediated growth⁸. AuNRs of different aspect ratio can be produced by working under controlled growth conditions following this approach^{9,10}. Many efforts have been made to control the characteristics of the synthesized NPs but, unfortunately, a mixture in terms of particle sizes and shapes is obtained in many cases. The principal drawback associated with the seeded growth approach to AuNRs synthesis with controllable aspect ratio is the formation of spherical NPs of different size as side products, as well as, shorter rods and platelets¹¹. The purification methods proposed are diverse with different potentials and limitations. The extraction and size selective precipitation methods allow separating AuNSs of different sizes but do not work for

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3 AuNRs shape separation¹². Nanoporous filter have been proposed for based shape
4 separation but there are problems when the short axis of nanorods and spheres have the
5 same diameter¹¹. AuNRs can be separated from small AuNSs by centrifugation but it is
6 not suitable for similar sizes of rods, spheres, and platelets^{13–15}. Moreover, it has been
7 proposed a surfactant assisted self-assembly as AuNRs shape separation technique, where
8 the centrifugation is used^{11,16}. In this case, the experimental conditions such as
9 concentration of the particle dispersion and a suitable particle stabilizer are very important
10 to achieve the separation. However, different stages of precipitation and redispersion
11 must be carried out to achieve an efficient separation and it is a very tedious
12 methodology.. Therefore, it is necessary the development of purification procedures
13 more efficient for the isolation of single size rods from a mixture of different rods and/or
14 spheres. Moreover, it is also very important the quality control of the synthesis and
15 purification procedures and they should be supported by appropriate analytical tools. ¹⁷.

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34 For the characterization of NPs, electron microscopy, such as transmission
35 electron microscopy (TEM) and scanning electron microscopy (SEM), among others, and
36 some spectroscopic techniques, such as Raman and X-rays, have been traditionally used.
37 However, all of them have different limitations mainly related to sample preparation, lack
38 of reproducibility and low sensitivity ^{18,19}. An outstanding alternative recently proposed
39 is the hyphenation of hydrodynamic separation techniques with sufficiently sensitive and
40 selective detectors ²⁰. These techniques have been mainly used for the separation of
41 AuNSs by size ^{21,22} and AuNRs like structured materials by aspect ratio ^{23–25} but, to the
42 best of our knowledge, there are very few applications based on the separation by shape
43 ^{26,27}. Size-exclusion chromatography ²⁶, and capillary electrophoresis ²⁷ were used with
44 diode-array detection in both cases. Among hyphenated systems, the separation by
45 asymmetric flow field flow fractionation (AF4) coupled to detection by inductively
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3 coupled plasma mass spectrometry (ICP-MS) is particularly interesting because the
4 advantages of AF4, in terms of resolving power and lack on interaction with the stationary
5 phase, are combined with the high sensitivity and selectivity of ICP-MS^{19,28–34}. In spite
6 of the potential advantages, as far as we know, AF4-ICP-MS has never been used for the
7 separation of different shapes of NPs with any detector.
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15 The aim of this work is the development of an analytical strategy based on the use
16 of AF4 coupled to ICP-MS to separate different AuNP shapes (AuNRs and AuNSs). The
17 experimental parameters that affect the separation of the AuNSs and AuNRs will be
18 optimized. The method will be applied for the quality control of the synthesis procedure
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30 **2. Materials and methods**

31 *2.1 Materials*

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36 All chemicals and reagents were of analytical grade. Solutions were prepared in
37 ultrapure water, which was obtained from Milli-Q® A10 TM water purification system
38 (Millipore Corporation, USA). AuNSs stabilized in 0.2 mM citrate (11.3±0.9 nm and
39 20.0±2.5 nm) and AuNRs in water (69.3±10.1 nm x 12±0.8 nm, 5.79 aspect ratio, and
40 103.2±10.5 nm x 18.5±2.1 nm, 5.63 aspect ratio) were purchased from NanoComposix
41 (USA). The concentrations of the commercial AuNSs, and AuNRs were estimated by
42 quantification of the total gold content by ICP-MS³⁰. The total gold concentrations were
43 51,886±2,183 ng mL⁻¹ (n=3) and 43,638±135 ng mL⁻¹ (n=3) for 10 nm and 20 nm AuNSs,
44 respectively; and 6,768±80 ng mL⁻¹ (n=3) and 4,322±56 ng mL⁻¹ (n=3) for 5.79 and 5.63
45 aspect ratio AuNRs, respectively.
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3 Rhodium, sodium dodecyl sulphate (SDS), $\text{NaH}_2\text{PO}_4 \cdot \text{H}_2\text{O}$ and $\text{Na}_2\text{HPO}_4 \cdot 12\text{H}_2\text{O}$
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5 were purchased from Inorganic Ventures (USA), Panreac (Spain) and Scharlab (Spain),
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7 respectively. Gold (III) chloride trihydrate ($\text{HAuCl}_4 \cdot 3\text{H}_2\text{O}$), tri-sodium citrate
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9 ($\text{Na}_3\text{C}_6\text{H}_5\text{O}_7$), sodium tetrahydridoborate (NaBH_4), cetyltrimethylammonium bromide
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11 (CTAB), ascorbic acid ($\text{C}_6\text{H}_8\text{O}_6$) were purchased from Sigma-Aldrich (USA).
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15 0.2 μm nylon filters (Agilent Technologies, Econofilter Nylon diameter 13 mm)
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17 and 0.1 μm membrane filters (Postnova, Germany) were used to filtrate the samples and
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19 the mobile phase, respectively.
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26 *2.2 Apparatus and instrumental features*

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28 The AF4 system was an AF2000 (Postnova Analytics, Germany). A membrane of
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30 regenerated cellulose (RC) with a molecular weight cut-off of 10 kDa was adapted as the
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32 accumulation wall. The trapezoidal channel was 27.5 cm in length and from 2 to 0.5 cm
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34 in width, and the spacer used was 350 μm thick. AF4 carrier solution was phosphate
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36 buffer (1 mM, pH 7) with the addition of SDS (0.01 %) and it was filtered through 0.1
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38 μm membrane filters and degassed.
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43 A Quadrupole ICP-MS Thermo X-SeriesII (Thermo Electron Corporation,
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45 Germany) equipped with a Meinhard nebulizer was employed. Rhodium ($15 \mu\text{g L}^{-1}$) was
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47 simultaneously aspirated during the ICP-MS data acquisition and it was applied to correct
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49 signal drifts. The raw data of the transient isotope signal for the different gold species and
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51 rhodium (internal standard) were exported from XSeries PlasmaLab software to
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53 OriginPro 2016 software, where they were processed.
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3 A spectrophotometer (model UV-3600, Shimadzu, Japan) was used to record
4 spectra at room temperature in the 190-1400 nm range, with resolution of 1 nm and 0.1
5 cm path length quartz cuvettes.
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10 TEM images were acquired on a JEOL JEM 1400 operating at an acceleration
11 voltage of 80 kV. Samples were prepared by drying a drop of 10 μL on a carbon-coated
12 copper grid.
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16 An advanced vortex mixer (Velp Scientifica, ZX3, Italy) was used to prepare
17 samples.
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23 24 25 26 *2.3 Procedure for the synthesis of 4.6 aspect ratio AuNRs*

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28 The 4.6 aspect ratio AuNRs were synthesized according to Jana et al.⁹. Briefly, a
29 seed solution (3.5 nm AuNSs) was prepared from 20 mL of a mixture of 2.5×10^{-4} M
30 HAuCl_4 and 2.5×10^{-4} M tri-sodium citrate, and 0.6 mL of ice cold 0.1 M NaBH_4 solution
31 in a conical flask under stirring. This seed solution was used within 2-5 h after
32 preparation. In a clean test tube, 10 mL of a growth solution (2.5×10^{-4} M HAuCl_4 and
33 0.1 M CTAB) was mixed with 0.05 mL of 0.1 M ascorbic acid. Finally, 0.025 mL of the
34 seed solution was added to the test tube, and after 5-10 min, the 4.6 aspect ratio AuNRs
35 are ready.
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52 **3. Results and discussion**

53 54 *3.1 Optimization of the AF4-ICP-MS method*

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56 Based on previous studies about AuNPs, a regenerated cellulose membrane
57 (MWCO of 10 KDa) and a 350 μm thick spacer were used³¹. When a membrane is used
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3 for the first time, it is common to do a pre-conditioning to avoid undesirable membrane-
4 particle interactions ^{35,36}. In our case, five consecutive injections of AuNSs were enough
5 for conditioning the new membrane. Moreover, according to the literature, a phosphate
6 buffer (1 mM NaH₂PO₄, 1 mM Na₂HPO₄ at pH 7) with the addition of 0.01 % SDS could
7 be appropriate as mobile phase for several reasons. The presence of monovalent cations
8 in the carrier can avoid the formation of aggregates and has no effect on the fractionation
9 times ³⁷; a correct selection of pH reduces the loss of particulate material by adsorption
10 on the membrane and avoids electrostatic attraction between the NPs and the membrane
11 ³²; and the addition of surfactants to the mobile phase increases the NPs stability and
12 contributes to minimize the possible aggregation or adsorption ³³.

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27 Once the dimensions of the channel, the type of membrane and the carrier
28 composition were selected, it was necessary to optimize several experimental parameters
29 related to the AF4 system, such as the focusing conditions and elution cross flow, and
30 injection/focusing time. A sample of 20 nm AuNSs (87 ng mL⁻¹) and 5.63 aspect ratio
31 AuNRs (104 ng mL⁻¹) prepared in the carrier solution from AuNPs commercially
32 available was used in the optimization study.

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41 One of the most important variables that affect the fractionation of NPs by AF4 is
42 the cross flow. The smallest particles require a great flow to be separated properly.
43 However, high flow rates lead to longer fractionation times and selective losses of the
44 largest particles in the sample. One option for samples with a wide range of particle size
45 is to apply gradients, that is, to reduce the cross flow over time. Therefore, different
46 gradients of the cross flow were evaluated and they are shown in Table S1. The injection
47 flow (0.20 mL min⁻¹), injection time (5 min) and cross and focus flow (0.50 and 0.80 mL
48 min⁻¹, respectively) were kept constant in the focusing step. The programs of gradient A,
49 B and C (Table S1) did not facilitate the separation (Figure S1, S2, and S3). However, the

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3 use of a gradient cross flow starting at 0.5 mL min^{-1} followed by a power decay (exponent
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5 1.5) for longer times (45 min) (Table 1) resulted in an appropriate separation between the
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7 void peak (first peak, 7.5 min) and that of the 20 nm AuNSs (second peak, 13.5 min), and
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9 between this and 5.63 aspect ratio AuNRs (third peak, 23 min) (Figure 1). The fraction
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11 of void peak could be due to a possible “memory effect” because of a release of NPs
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13 during carrier injection or the presence of a small particles fraction in the AuNPs
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15 commercially available³⁴. In order to evaluate if the injection time has an effect on the
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17 separation of NPs, the injection/focusing time was modified from 2.5 to 7.5 min. As
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19 illustrated in Figure S4, longer injection times produced a shift to longer fractionation
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21 times and a decrease in the fraction of void peak. Besides, it was observed that the
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23 intensity of AuNSs and AuNRs peaks was high for an injection time of 5 min, so it was
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25 selected for further experiments (Table 1).
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32 The AF4 theory can be used to estimate the hydrodynamic diameters of the AuNSs
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34 and AuNRs from their retention times in the fractograms (Figure 1). The retention time
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36 of the AuNSs (13.5 min) can be related to the diffusion coefficient according to Runyon
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38 et al. equation ($t_r = (w^2/6D) * \ln(1 + (V_c/V))$), retention time (t_r), channel thickness (w),
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40 diffusion coefficient (D), cross flow rate (V_c), and flow rate exiting through the channel
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42 outlet (V)²⁵, and subsequently to the hydrodynamic diameter (Stokes-Einstein equation:
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44 $d_h = kT/3\pi\eta D$, hydrodynamic diameters (d_h), Boltzmann constant ($k = 1.38 \times 10^{-23} \text{ kgm}^2\text{s}^{-2}\text{K}^{-1}$),
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46 temperature (T), viscosity ($\eta = 0.000891 \text{ kgm}^{-1}\text{s}^{-1}$), and diffusion coefficient (D)), which
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48 was 28 nm. However, Beckett et al. established that the retention time does not depend
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50 only on the diffusion coefficient, since a certain dependence of the shape would be
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52 expected³⁸. In the case of AuNRs, the retention time (23 min) is principally dependent
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54 on the translational diffusion coefficient (not of the rotational diffusion coefficient)²⁵,
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56 which can be determined according to the El Hadri et al. equation ($D_{t-AF4} = V_c w^2 t_0 / 12 V_0 t_r$),
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3 translational diffusion coefficient (D_t), cross flow rate (V_c), channel thickness (w), void
4 time (t_0), void volume (V_0), and retention time (t_r)²³. Using the Stokes-Einstein equation
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6 and taking into account that D_t corresponding to the cylinder model (aspect ratio between
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8 2 to 20) equals the D ²³. The equivalent sphere diameter for the AuNRs was 66.3 nm. As
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10 expected, the hydrodynamic diameter of the AuNRs (66.3 nm) is greater than the AuNSs
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12 (28 nm) and this justifies the separation found by AF4.
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18 Several experiments with AuNRs of different aspect ratios (5.63 and 5.79) and
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20 AuNSs of different sizes (10 and 20 nm), were carried out to demonstrate the strength of
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22 the method as a whole. As illustrated in Figure S5, the separations were satisfactory and
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24 the retention times changed with different aspect ratios (Figure S5, black line *versus* grey
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26 line) and sizes (Figure 1 *versus* Figure S5). Besides, different concentrations of 10 nm
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28 AuNSs (27, 54, 108, and 216 ng mL⁻¹) were spiked to samples containing 5.63 aspect
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30 ratio AuNRs (60 ng mL⁻¹). The samples were analysed and their fractograms are shown
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32 in Figure 2. It was possible to verify that the intensity of the AuNRs peak remained
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34 constant while the intensity of the AuNSs peak increased. This demonstrates the
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36 feasibility of the proposed method to separate mixtures of AuNRs and AuNSs at different
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38 concentration ratios.
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45 3.2. Study by AF4-ICP-MS and complementary techniques of AuNRs synthesis 46 procedure 47

48 The optimized AF4-ICP-MS method will be used for the quality control of
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50 products obtained after the synthesis of 4.6 aspect ratio AuNRs using a seeding growth
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52 method⁹. Samples (dilution 1/100) obtained in the synthesis were analyzed and a
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54 fractogram is shown in Figure 3a. A first peak (7.5 min) corresponding to the fractionation
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56 time of the void peak is observed (Figure 3a). Its high intensity could be related to
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3 impurities of the synthesis procedure, maybe small AuNSs (3.5 nm or lesser) of the seed
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5 solution.
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8 The second peak (Figure 3a) at 19 min (estimated diameter of 54.7 nm) is close
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10 to the AuNRs commercially available with an aspect ratio of 5.63 (Figure 1). This peak
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12 corresponds to the expected dimensions of the AuNPs according to the synthesis
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14 conditions and the low intensity could indicate that the yield was limited. The response
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16 factor method was used to quantify the concentration of AuNRs from the peak area in the
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18 registered fractogram. The 5.63 aspect ratio AuNRs commercial solution (104 ng mL^{-1})
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20 was the reference selected for the quantification. The estimated concentration of the
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22 AuNRs in the synthesized sample was $3,400 \text{ ng mL}^{-1}$.
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27 Moreover, it is necessary to know the fractionation time and intensity of the
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29 AuNPs commercially available in the synthesis medium for an adequate comparison.
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31 Therefore, samples obtained in the synthesis procedure were spiked independently with
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33 commercially available 20 nm AuNSs (873 ng mL^{-1}) or 5.63 aspect ratio AuNRs ($1,040$
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35 ng mL^{-1}), and they were analyzed (Figure 3b). The fractionation times of the
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37 commercially available AuNPs spiked (Figure 3b) were similar to those previously
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39 obtained in the carrier solution (Figure 1). This fact confirms that the fractionation times
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41 do not change in the synthesis medium and that the second peak found in the Figure 3a
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43 could be the 4.6 aspect ratio AuNRs similar to the commercially available AuNRs (5.63
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45 aspect ratio). However, the intensity of the commercially available AuNPs in the
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47 synthesis medium are lower than the expected. Low recovery values cannot be attributed
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49 to undesirable membrane-particle interactions because the membrane had been
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51 previously stabilized and will be further investigated using the information given in the
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53 fractograms.
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3 The third peak (Figure 3a) at 57.5 min represents AuNRs with an aspect ratio
4 greater than 5.63 or aggregates/agglomerates of AuNPs, which was not expected
5 according to the used synthesis. When the samples obtained in the synthesis procedure
6 were spiked independently with both commercially available AuNPs (Figure 3b), the
7 intensity of the second peak was lower than expected, while the third peak was even
8 higher than in Figure 3a. These changes could indicate the possible formation of
9 aggregates/agglomerates of NPs.
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20 The different spectroscopic absorption characteristics of the AuNSs and AuNRs
21 were exploited to get a closer insight into these processes. The absorption spectra of a
22 sample from the synthesis procedure of 4.6 aspect ratio AuNRs, as well as the
23 commercially available AuNSs and AuNRs were recorded (Figure 4). As expected, the
24 commercially available 20 nm AuNSs has an absorption maximum at 500 nm (Figure 4,
25 grey line), while the commercially available 5.63 aspect ratio AuNRs has two, the first
26 one at 500 nm and the second one at 950 nm (Figure 4, dash line). However, the sample
27 from the synthesis has a major band at 500 nm (Figure 4, black line). This band indicate
28 the presence of AuNSs, probably as impurities of the seed solution in the synthesis
29 procedure. Moreover, the lack of a major band in the near-infrared region indicates that
30 there is not a significant amount of AuNRs. These results agree with those previously
31 reported by AF4-ICP-MS (Figure 3a).
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48 In addition, the TEM analysis of the sample from the synthesis procedure of 4.6
49 aspect ratio AuNRs (dilution 1/100) was carried out (Figure 5). A mixture of AuNRs of
50 different aspect ratios, AuNSs and other shapes, was found. The TEM image confirmed
51 the existence of AuNRs with the expected aspect ratio and also showed the presence of
52 other much higher ones, which confirms the information obtained by AF4-ICP-MS.
53 However, the TEM image cannot show the existence of small AuNSs (3.5 nm or lesser)
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3 of the seed solution, probably, because the size is too small to be detected by the
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5 instrument used.
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8 Problems related to the synthesis procedure for small AuNRs (4.6 aspect ratio)
9 and difficulties for purification have been also reported by other authors ⁹. Therefore, it
10 is very important to perform an adequate quality control and the developed method based
11 on AF4-ICP-MS has demonstrated its capability for this purpose. Moreover, AF4 can be
12 useful in the fractional collection mode to separate the particle mixture into monodisperse
13 NPs. This approach has been already used by Runyon et al. to separate AuNRs of different
14 aspect ratio using AF4-UV-vis. Moreover, they collected the fractions which were
15 analyzed by UV-vis and TEM, and one of them was a mixture of AuNRs and AuNSs ²⁵.
16 Therefore, this potential should be explored in further studies for purification and quality
17 control.
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35 **4. Conclusions**

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38 A methodology based on AF4 hyphenated to ICP-MS has been proposed to
39 separate AuNPs of different shapes, namely AuNSs and AuNRs. After the optimization
40 of the adequate separation conditions, its applicability for quality control of synthesis
41 procedures of AuNRs has been demonstrated. In combination with UV-vis molecular
42 absorption spectroscopy and TEM as complementary techniques, the presence of
43 significant impurities has been pointed out.
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52 The development of hyphenated techniques, such as the proposed in this work,
53 for the separation of NPs by shape is of major interest because this characteristic is very
54 important for their applications in different fields. Further experiments should be carried
55 out to evaluate the capabilities of this approach as a tool for purification purposes.
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Supplementary Material

Supplementary data associated with this article is provided.

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Legends to figures

Figure 1. AF4-ICP-MS fractogram of a mixture containing commercially available 20 nm AuNSs (87 ng mL^{-1}) and 5.63 aspect ratio AuNRs (104 ng mL^{-1}). Optimal cross flow program Table 1 (dash line). Carrier: 0.01 % SDS, 1 mM NaH_2PO_4 , 1 mM Na_2HPO_4 , pH 7.

Figure 2. AF4-ICP-MS fractogram of a mixture containing commercially available 10 nm AuNSs at different concentrations: 27 ng mL^{-1} (black line), 54 ng mL^{-1} (grey line), 108 ng mL^{-1} (black dash line), and 216 ng mL^{-1} (grey dash line), and 5.63 aspect ratio AuNRs (60 ng mL^{-1}). Optimal cross flow program Table 1. Carrier: 0.01 % SDS, 1 mM NaH_2PO_4 , 1 mM Na_2HPO_4 , pH 7.

Figure 3. AF4-ICP-MS fractograms of a sample obtained in the synthesis procedure of 4.6 aspect ratio AuNRs (dilution 1/100): a) directly, b) spiked with commercially available 20 nm AuNSs (873 ng mL^{-1}) (grey line) or with 5.63 aspect ratio AuNRs ($1,040 \text{ ng mL}^{-1}$) (dash line). Optimal cross flow program Table 1. Carrier: 0.01 % SDS, 1 mM NaH_2PO_4 , 1 mM Na_2HPO_4 , pH 7.

Figure 4. Absorption spectra of a sample obtained in the synthesis procedure of 4.6 aspect ratio AuNRs (black line), the commercially available 20 nm AuNSs ($43,638 \text{ ng mL}^{-1}$) (grey line) and 5.63 aspect ratio AuNRs ($4,332 \text{ ng mL}^{-1}$) (dash line).

Figure 5. TEM image of a sample obtained in the synthesis procedure of 4.6 aspect ratio AuNRs (dilution 1/100).

Table 1. Optimum instrumental conditions for the AF4-ICP-MS system.

| AF4 | | | | |
|---|--|--|-------------------------------------|--|
| Channel thickness (μm) | 350 | | | |
| Membrane type | Regenerated cellulose; cut-off, 10 kDa | | | |
| Injection loop (μL) | 50 | | | |
| Carrier liquid composition | 0.01 % SDS, 1mM NaH_2PO_4 , 1mM Na_2HPO_4 , pH 7 | | | |
| Focus flow (mL min^{-1}) | 0.8 | | | |
| Detector flow (mL min^{-1}) | 0.5 | | | |
| Cross flow program | Time (min) | Regime | Cross flow (mL min^{-1}) | |
| Injection/focusing | 5 | Injection flow, 0.2 mL min^{-1} | 0.5 | |
| Elution cross flow | 45 | Power (Exponent 1.5) | 0.5 to 0 | |
| | 10 | Constant | 0 | |
| ICP-MS | | | | |
| RF-Power (KW) | 1.4 | | | |
| Plasma gas flow rate (L min^{-1}) | 15 | | | |
| Carrier gas flow rate (L min^{-1}) | 0.98 | | | |
| Nebulizer flow rate (L min^{-1}) | 0.9 | | | |
| Auxiliary gas flow rate (L min^{-1}) | 1 | | | |
| Spray chamber | Scott type | | | |
| Isotopes monitored | ^{197}Au , ^{103}Rh | | | |
| Dwell time (ms) | 100 | | | |

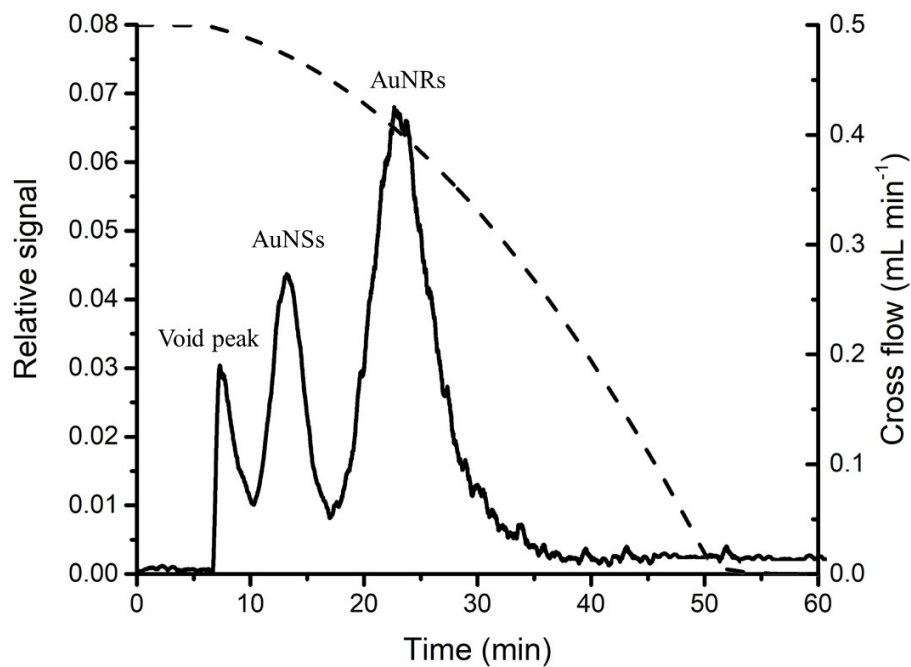


Figure 1. AF4-ICP-MS fractogram of a mixture containing commercially available 20 nm AuNSs (87 ng mL⁻¹) and 5.63 aspect ratio AuNRs (104 ng mL⁻¹). Optimal cross flow program Table 1 (dash line). Carrier: 0.01 % SDS, 1 mM NaH₂PO₄, 1 mM Na₂HPO₄, pH 7.

249x190mm (120 x 120 DPI)

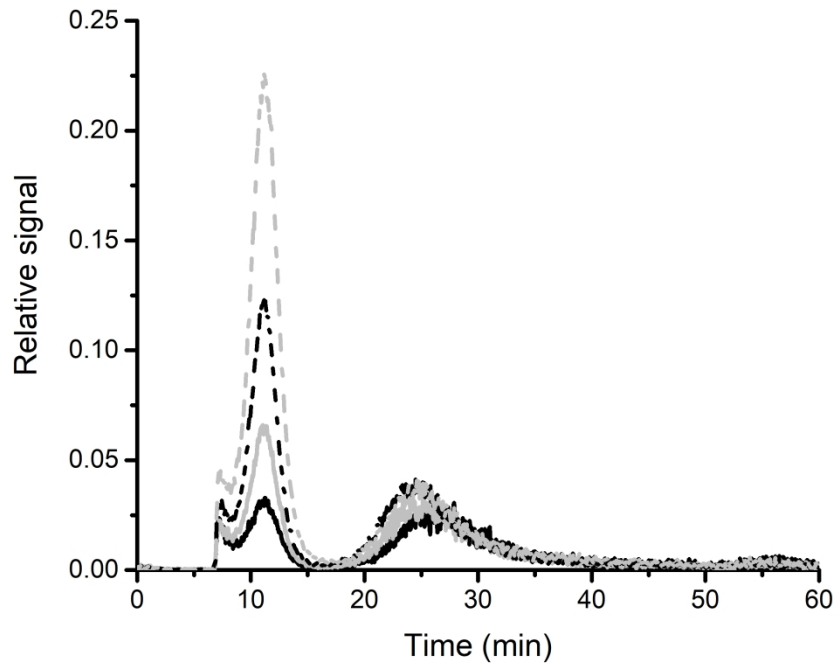


Figure 2. AF4-ICP-MS fractogram of a mixture containing commercially available 10 nm AuNSs at different concentrations: 27 ng mL⁻¹ (black line), 54 ng mL⁻¹ (grey line), 108 ng mL⁻¹ (black dash line), and 216 ng mL⁻¹ (grey dash line), and 5.63 aspect ratio AuNRs (60 ng mL⁻¹). Optimal cross flow program Table 1. Carrier: 0.01 % SDS, 1 mM NaH₂PO₄, 1 mM Na₂HPO₄, pH 7.

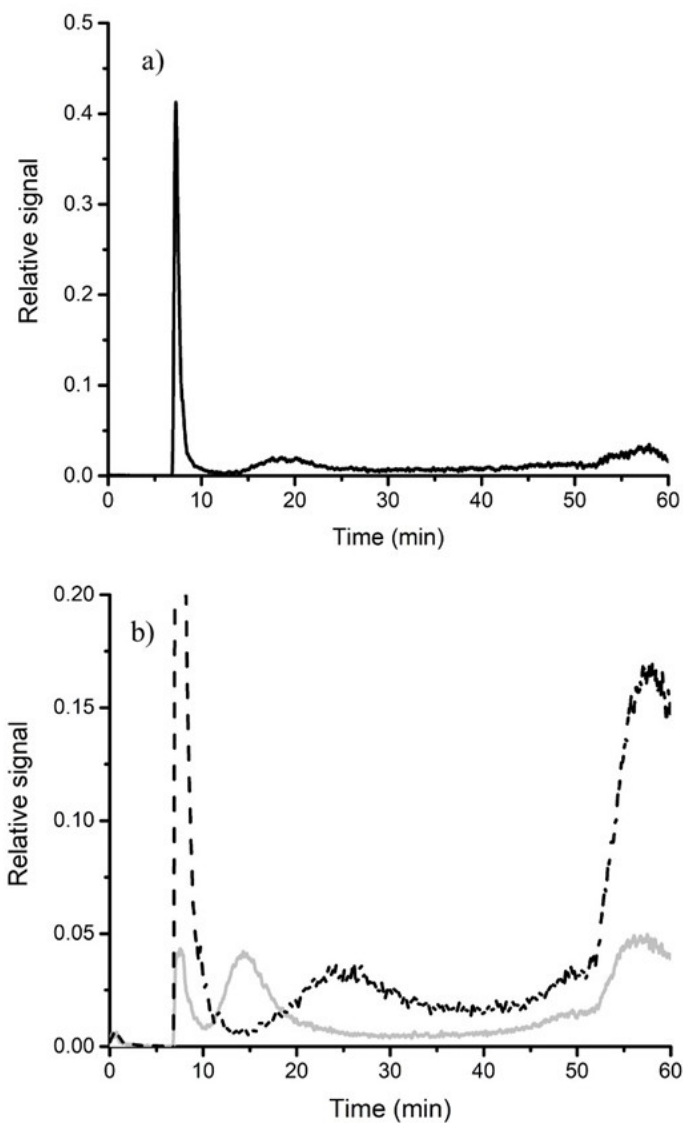


Figure 3. AF4-ICP-MS fractograms of a sample obtained in the synthesis procedure of 4.6 aspect ratio AuNRs (dilution 1/100): a) directly, b) spiked with commercially available 20 nm AuNSs (873 ng mL⁻¹) (grey line) or with 5.63 aspect ratio AuNRs (1,040 ng mL⁻¹) (dash line). Optimal cross flow program Table 1. Carrier: 0.01 % SDS, 1 mM NaH₂PO₄, 1 mM Na₂HPO₄, pH 7.

131x190mm (120 x 120 DPI)

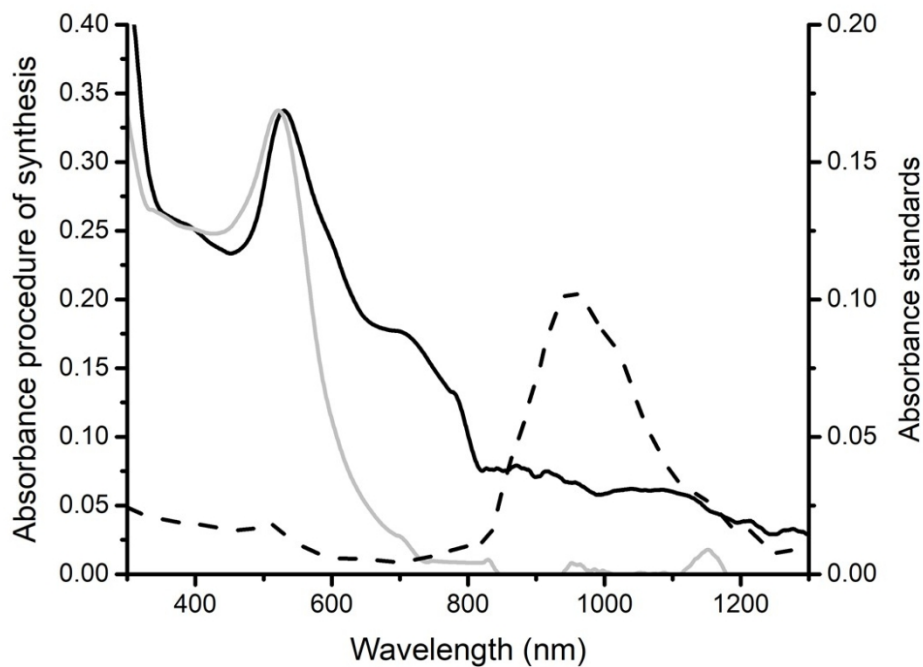
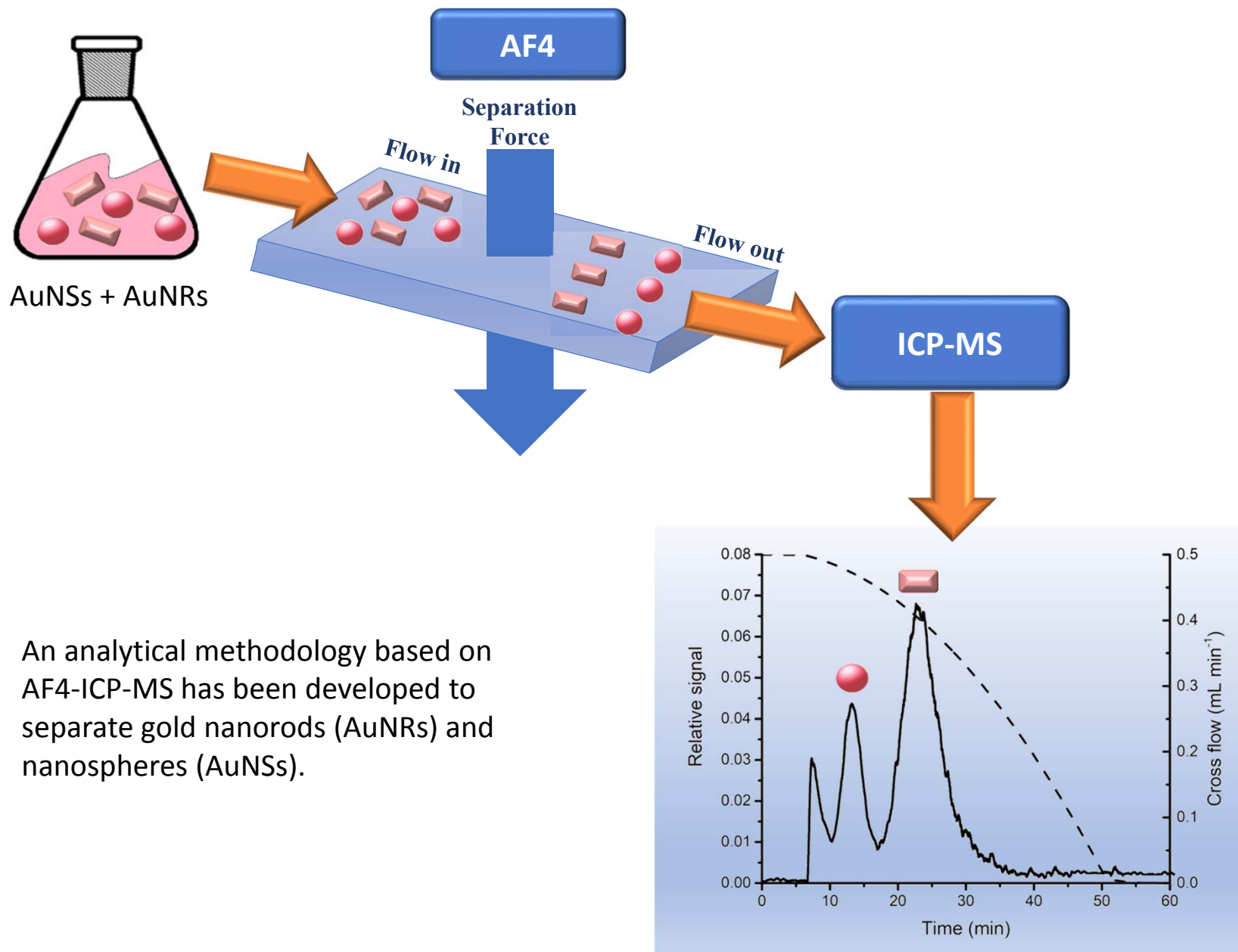


Figure 4. Absorption spectra of a sample obtained in the synthesis procedure of 4.6 aspect ratio AuNRs (black line), the commercially available 20 nm AuNSs (43,638 ng mL⁻¹) (grey line) and 5.63 aspect ratio AuNRs (4,332 ng mL⁻¹) (dash line).

248x190mm (120 x 120 DPI)



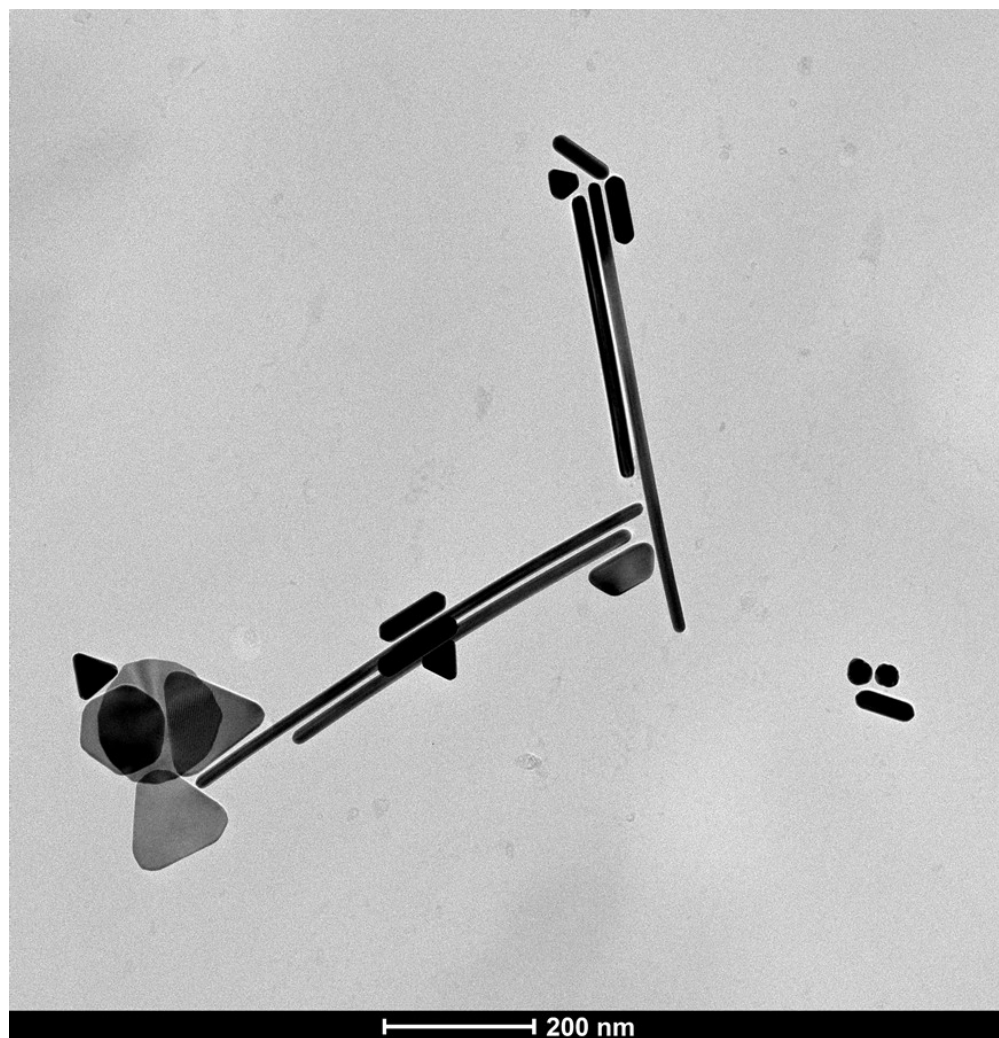


Figure 5. TEM image of a sample obtained in the synthesis procedure of 4.6 aspect ratio AuNRs (dilution 1/100).

184x190mm (120 x 120 DPI)