ACADEMIA ROMÂNĂ
Revue Roumaine de Chimie
http://web.icf.ro/rrch/

Rev. Roum. Chim.,
2015, *60*(7-8), 721-726

Dedicated to Professor Valer Farcasan on the occasion of his 95th anniversary

GREEN SYNTHESIS AND CHARACTERIZATION OF GOLD AND SILVER NANOPARTICLES

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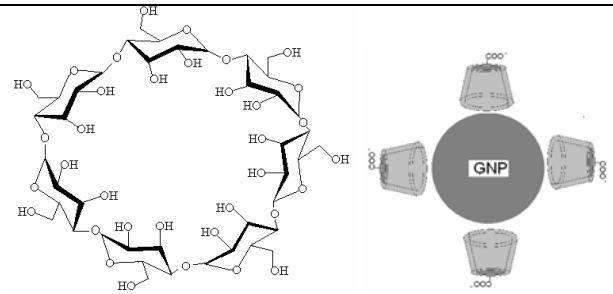
Received January 16, 2015

Gold nanoparticles (GNPs) and silver nanoparticles (SNPs) are very attractive materials for nanotechnology, nanobiology and nanomedicine. We employ β-cyclodextrin (βCD) both as reducing and stabilizing agent for the preparation of GNPs and SNPs aqueous dispersions, to be used in biological applications. $HAuCl_4$ and respectively $AgNO_3$ were reduced with βCD in alkaline medium at room temperature. Reactions were monitored by measuring the intensity of the characteristic surface plasmon resonance (SPR) absorption band at 520 nm for GNPs, and at 404 nm for SNPs. The size of particles was determined from TEM images. Particularly, the average diameter of GNPs is 7.6 nm and for SNPs is 13.1 nm. The dispersions remained stable for at least a year after preparation. The nanoparticles were further characterized by AFM imaging, zeta potential and dynamic light scattering measurements, as well as by FT-Raman spectra. Their stability against acid (HCl) and saline (NaCl) solutions was also investigated.



INTRODUCTION

Gold nanoparticles (GNPs) and silver nanoparticles (SNPs) are very attractive materials for nanotechnology, nanobiology or nanomedicine,[1-7] because of their unique physical and chemical properties that originate from the high surface area to volume ratio. Both GNPs and SNPs are essentially to possess high chemical stability and suitable biocompatibility to lead them to promising biological and biomedical applications. In particular, it is important that the prepared colloidal solutions should not contain toxic substances and in consequence, green synthesis[8,9] is preferred. In addition, the particles size distribution is required as narrow as possible and small sizes are advantageous. Also, the GNPs and SNPs colloidal systems must be stable against aggregations of nanoparticles. Further, GNPs and SNPs should possess proper surface functionalization to allow the conjugation of nanoparticles with biologically active molecules. Certainly, the development of facile, eco-friendly and inexpensive preparation methods is of current interest and this research on GNPs and SNPs broadens their potential applications in biomedicine, including drug delivery systems.

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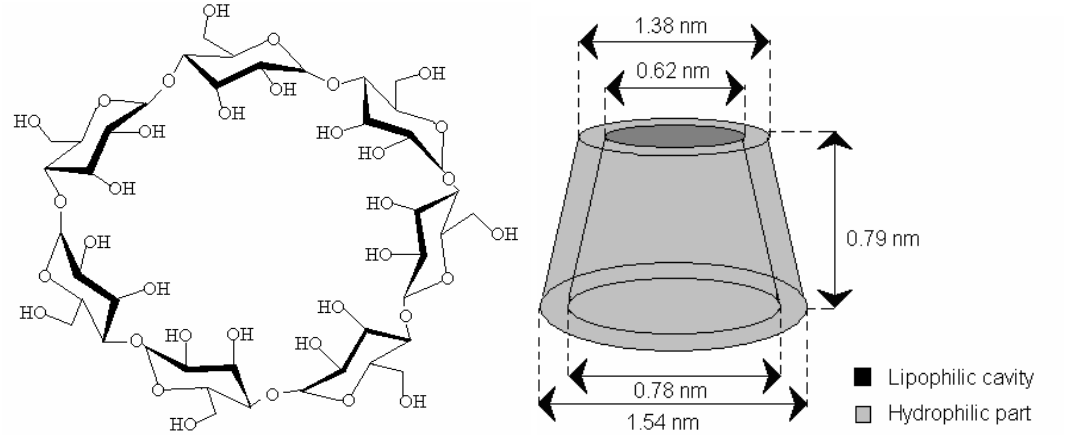


Scheme 1 – Structure and geometric sizes of β-cyclodextrin.

β-Cyclodextrin (βCD) is a carbohydrate produced by enzymatic alteration of starch and is frequently used as carrier for bioactive molecules.[10-12] It is a cyclic oligosaccharide (Scheme 1) with seven glucose residues, linked by α (1-4) glycosidic bonds.[13-15] It can be viewed as a toroid with the sizes given in Scheme 1. The primary hydroxyl (-OH) groups are oriented to the exterior of its smaller opening, while the secondary hydroxyls are located at the larger one. Thus the exterior of the toroid is hydrophilic, while the interior can be considered hydrophobic with respect to the aqueous medium (lipophilic).

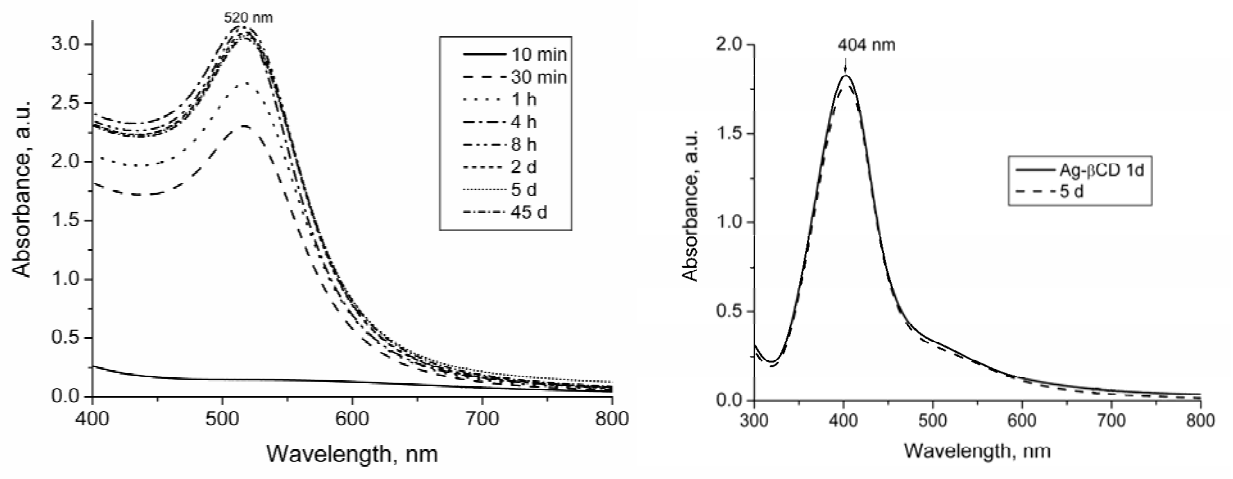
Cyclodextrins or their derivatives were sometimes used for the control of size distribution and as stabilizers for GNPs or for SNPs, obtained by chemical reduction with citrate or borohydride,[16-20] and a more uniform size distribution and lower diameters of the nanoparticles were observed. A similar effect was ascertained for the presence of cyclodextrins in the process of laser ablation for producing very small GNPs.[21]

Furthermore, the use of cyclodextrins as reducing agents for the preparation of core-shell bimetallic particles of Au and Ag was investigated.[22] The reduction of $HAuCl_4$ with α-CD was used for the production of GNPs for applications in catalysis and SERS,[23] while βCD capped GNPs were applied in the selective detection of Pb^{2+} ions.[24] βCD was as well used to obtain a GNPs–βCD–graphene-modified electrode for voltammetry.[25]

While there are reports on both GNPs and SNPs capped with βCD, a little is known about the uses of βCD as a reducing agent and simultaneously as a stabilizing agent in the chemical preparation of GNPs or of SNPs in aqueous media. This study is aimed to employ βCD as a reducing agent for the controlled green chemical synthesis of both GNPs and SNPs in aqueous media, thus, eliminating any hazardous reducing agent as well as the problems of its disposal. In addition, βCD is also utilized as a stabilizing agent to protect both GNPs and SNPs from their aggregation and further regulating their surface properties and making them able to form various hybrid nanoconjugates with diverse biomolecules for biological and biomedical applications.

RESULTS AND DISCUSSION

The characteristic absorption band of surface plasmon resonance (SPR) for GNPs with the maximum at about 520 nm appeared some minutes after the mixing of the $HAuCl_4$ and βCD solutions at pH 11, and its intensity increased gradually (Fig. 1a), while the color of the solution became dark red. After 4 hours at room temperature the maximum intensity was attained. In the mixture of $AgNO_3$ and βCD solutions, at pH 9, the yellow color, denoting the formation of SNPs appeared at once, and continued to intensify for an hour. In the UV-Vis spectrum, the characteristic SPR absorption band had its maximum at 404 nm (Fig. 1b). Both GNPs and SNPs systems were stable more than one year after preparation. The stability of the GNPs system against acid and salt solutions was tested. Adding a 0.1 M HCl solution or a 0.5 M NaCl solution, to the GNPs colloidal solution, causes the aggregation and sedimentation of the GNPs.



a. b.

Fig. 1 – Time evolution of the SPR absorption band for GNPs (a), and SNPs (b).

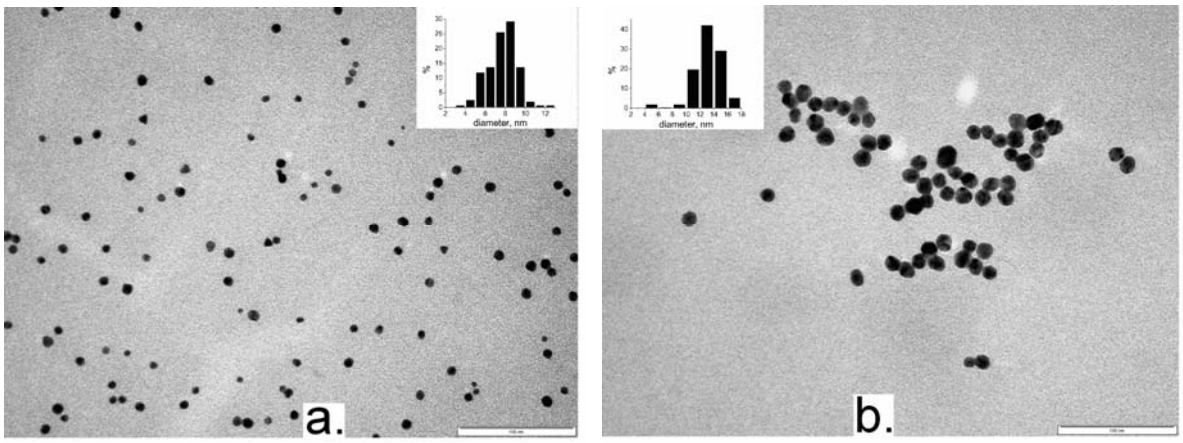


Fig. 2 – TEM images of GNPs (a) and SNPs (b). The bars in TEM images correspond to 100 nm
Inserts are the histograms of size distribution.

A representative TEM image of the GNPs is given in Fig. 2a, while a particular TEM image for SNPs is shown in Fig. 2b. TEM images illustrate that both gold and silver nanoparticles are generally of spherical shape, but some polyhedral forms (triangular, hexagonal) are also observed. In the case of SNPs, a trend to build nano swarms of loosely bonded particles is observed.

The diameters of a great number (hundreds) of particles were measured on the TEM images, and histograms providing the size distribution of GNPs and SNPs are inserted in Fig. 2a and 2b, respectively. The size distribution is quite narrow; for GNPs the average size (diameter) of the particles is 7.6 ± 1.5 nm, while the SNPs are larger, the average diameter being 13.1 ± 2.1 nm. By dynamic light scattering (DLS) a somewhat higher value for the diameters of the particles is found, namely 10.1 nm for GNPs, respectively 14.7 for SNPs. This could be explained by the fact that DLS does not distinguish between the metal nanoparticles and the organic molecules coating them, while in the TEM images the dark (black) spots correspond to the metal particles, which have a much higher density than the organic coating.

The zeta potential measured for the GNPs was -23.4 mV, while ±30 mV is considered as the limiting value assuring the electrostatic stability of colloid systems.[26] For SNPs the zeta-value found was even less negative: -12.3 mV. Therefore the high stability observed for the gold and silver nanoparticles obtained by reduction with βCD cannot be primarily assigned to electrostatic repulsions among particles as preventing factor against the coagulation of the colloidal systems, but rather to the effect of the βCD coating layer on the surface of nanoparticles.

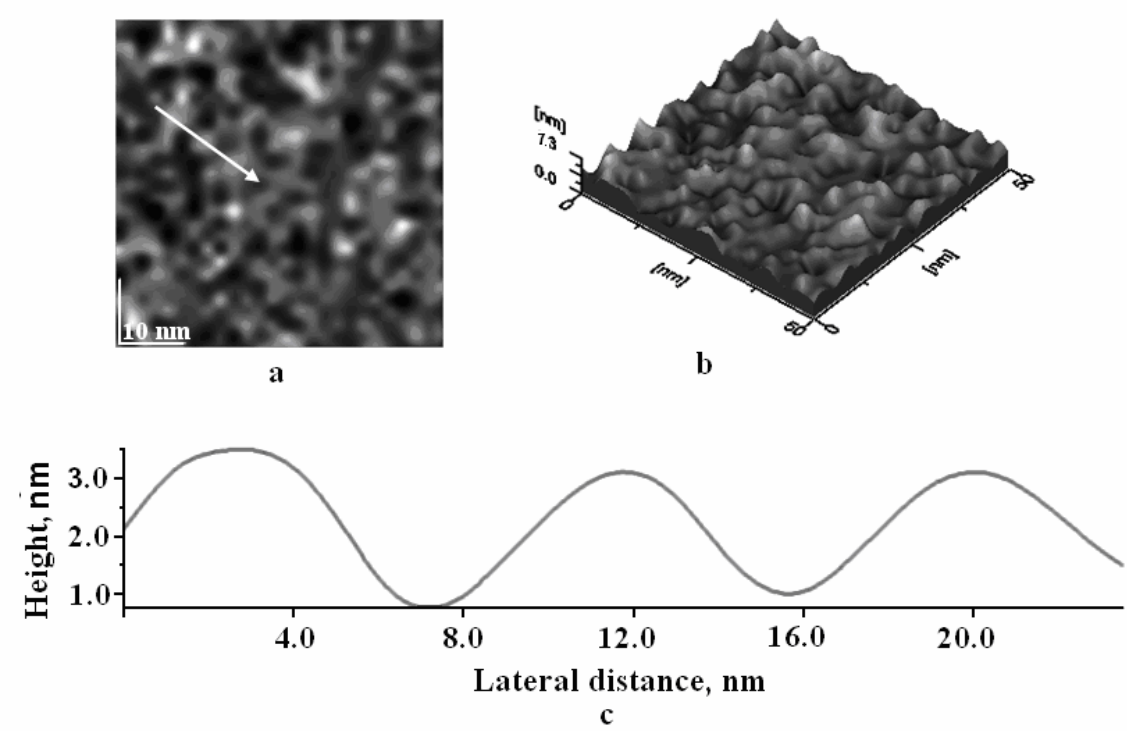


Fig. 3 – AFM images of GNPs assembled on glass: (a) 2D- topography; (b) 3D – topography; (c) profile of the cross section along the arrow in panel a; scanned area 50 nm x 50 nm.

As an exemplification of AFM images, we present topographical images and a plot of the film surface contour in a cross section for GNPs in Fig. 3. It should be noted that morphologies (self assemblies) observed by AFM might be rather different to those present in colloidal solutions, as a consequence of drying and surface effects. Nevertheless, AFM images give a valuable illustration on the structuration of adsorbed gold nanoparticles on glass surface. Therefore, from the profile of the cross section (Fig. 3c), the size of GNPs is seen to be about 6 ± 2 nm, which is substantially consistent with that found from TEM images. It is to be noted that, similar results were also found with SNPs assembled on glass surface investigated by AFM, and the size of silver nanoparticles of about 12 ± 2 nm is in substantial agreement with TEM investigations.

The FT Raman spectra of the 10^{-2} M βCD solution and of the same solution after the reaction with $HAuCl_4$ and the formation of the GNPs are represented in Fig. 4. The spectra are quite similar, no major differences could be revealed. Therefore, direct Au-O interactions between the gold atoms and the 21 hydroxyl groups of βCD are not probable. On the other hand, the inclusion of gold nanoparticles in the cavities of the βCD molecules is impossible, since the inner diameter of such a cavity is only 0.78 nm (Scheme 1). The stabilization of the GNPs in presence of βCD molecules could be ascribed to hydrophobic interactions between the more hydrophobic part of the larger opening of the βCD toroid molecules (Scheme 1) and GNPs. In other words, GNPs might be located on the top of such βCD cavities, which would prevent the aggregation of GNPs.[16]

We can assume that primary hydroxyl groups of βCD are oxidized to –COOH groups, ionized in alkaline medium as $–COO^-$.[23, 24] The reactions occurring during the reduction could be written as:

$$4\ NaAuCl_4 + 3RCH_2OH + 15\ NaOH \rightarrow$$

$$\rightarrow 4\ Au + 3\ RCOONa + 16\ NaCl + 12\ H_2O$$

$$4\ AgNO_3 + RCH_2OH + 5\ NaOH \rightarrow$$

$$\rightarrow 4\ Ag + RCOONa + 4\ Na\ NO_3 + 4\ H_2O$$

The $–COO^-$ groups contribute to the negative charge of the GNPs, as found by the zeta potential measurements, and their mutual repulsion adds to the stability of the system.[23] The presence of NaCl or $NaNO_3$ in the solution, arisen from the reactions, does not destabilize the system and assures a ionic strength of the solution which facilitates the conjugation with different negatively charged biologically active molecules.

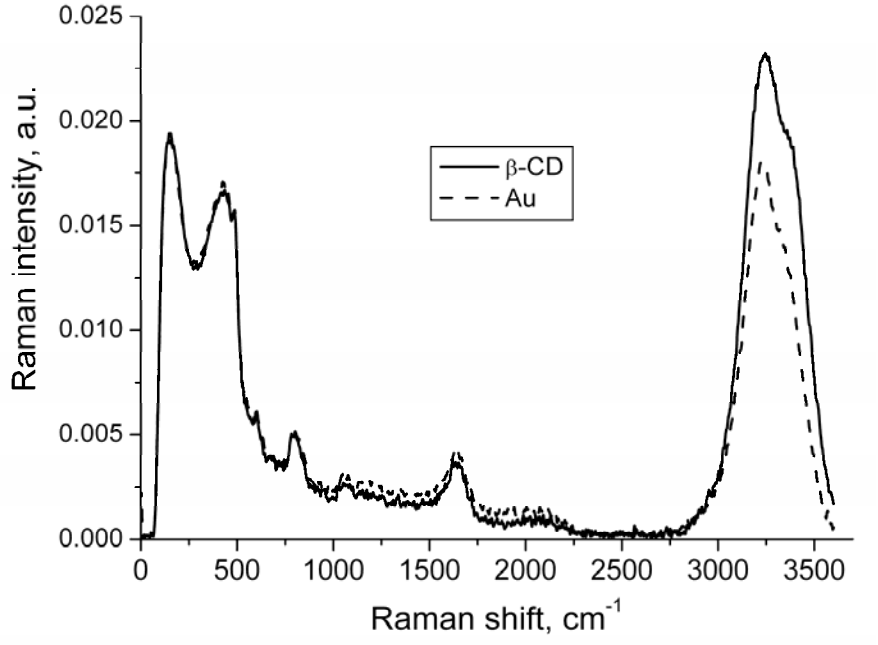


Fig. 4 – FT Raman spectra of a 10^{-2} M βCD solution (full line) and of final dispersion with gold nanoparticles obtained in 10^{-2} M βCD medium (dotted line).

EXPERIMENTAL

Materials. All chemicals used were of analytical grade or of the highest purity available and were used as received without further purification. Hydrogen tetrachloroaurate(III) trihydrate ($HAuCl_4 \cdot 3H_2O$), silver nitrate ($AgNO_3$), sodium hydroxide (NaOH) and potassium carbonate (K_2CO_3) pro analysis were purchased from Merck, Darmstadt, Germany. β-Cyclodextrin (βCD) was purchased from Sigma-Aldrich Chemical, Corp., Milwaukee, WI, USA. Aqueous solutions were prepared with double distilled water, which was further deionized (i.e., the resistivity of 18 Mohm·cm) in Elgastat water purification system.

Synthesis of GNPs. In order to prepare 100 mL colloidal solution with a 10^{-3} M gold content, a 1% $HAuCl_4 \cdot 3H_2O$ solution (3.9 mL) was mixed at room temperature with a 10^{-2} M aqueous βCD solution (96.1 mL), and the pH was adjusted to 11, by adding solid NaOH. The mixture was stirred vigorously for 1 hour.

Synthesis of SNPs. A colloidal solution with 10^{-3} M silver content was prepared by mixing 10^{-2} M βCD and 10^{-2} M $AgNO_3$ aqueous solutions in the 9:1 volume ratio, at room temperature, and the pH was adjusted to 9 (with K_2CO_3). The colloidal SNPs solution was diluted 5-fold with ultrapure water to $2 \cdot 10^{-4}$ M silver concentration.

The reaction progress was monitored by measuring the UV-Vis spectrum from time to time, for each of the two syntheses for GNPs and SNPs. The pH value of both GNPs and SNPs colloidal solutions decreased in time, to pH 6 eventually.

Methods. Optical absorption spectra were obtained using a Jasco UV-Vis V-650 spectrophotometer with 10 mm path length quartz cuvettes in the 190 – 900 nm wavelengths range.

Zeta potential and dynamic light scattering (DLS) measurements were performed using the Malvern Zetasizer Nano-ZS90, on the colloidal gold and silver solutions.

Raman FT spectra were obtained with the FRA 106/S FT-Raman Module attached to Bruker EQUINOX 55; an Nd:YAG laser was used (wavelength 1064 nm) and a liquid nitrogen cooled germanium detector (D418-T). The laser power was 350 mW, the resolution 4 cm^{-1}, the number of scans: 500.

The samples were observed with a *transmission electron microscope* (TEM, JEOL – JEM 1010). The nanoparticles suspensions (approximately 7 μL for each sample) were deposited on the 300 mesh electrolytic copper grids, coated with a carbon layer, and adsorbed for 1 min. The excess solution was removed with filter paper and the samples were air dried. TEM images have been recorded with JEOL standard software.

Atomic force microscopy, AFM, imaging was obtained using the AFM JEOL 4210 equipment, operated in tapping mode,[6,7] using standard cantilevers with silicon nitride tips (resonant frequency in the range of 200-300 kHz, spring constant 17.5 N/m). Images were performed on the nanostructured films of GNPs and of SNPs, obtained by vertical adsorption from their aqueous dispersions on glass plates for 10s and dried out in air. Different areas from 10 μm x 10 μm to 0.05 μm x 0.05 μm were scanned on the same film. The AFM images (2D- and 3D- topographies) and the cross-section profile in the film along a selected direction were processed by the standard AFM procedures.

CONCLUSIONS

We found that βCD can be used as a soft reducing agent for the reduction of Au(III) to Au(0) and of Ag(I) to Ag(0), and at the same time, it is a good stabilizing agent for the aqueous dispersions of the obtained nanoparticles. The reduction takes place in alkaline medium at room temperature, and the resulted system of

nanoparticles presents a narrow size distribution and a high stability in time. These results provide a reproducible and controlled green synthesis of GNPs and SNPs capped with βCD in aqueous dispersions. The nanoparticles agglomerate only at low pH and high saline concentration. This high stability cannot be reduced to the presence of negative charges on the GNPs and SNPs, but must imply the presence of a βCD coating layer on their surface, due mainly to hydrophobic interactions. However, the interaction among –OH groups on βCD and the surface of GNPs or SNPs cannot be ruled out completely. This preparation method for both GNPs and SNPs is simple, takes place at room temperature, and does not imply harmful chemicals, so both the GNPs and SNPs should be appropriate for potential biological and biomedical applications. Our preliminary results show that the βCD coating layer on metallic nanoparticles does not prevent the interaction of metallic nanoparticles with biological active compounds.

Acknowledgements: We acknowledge the Executive Agency for Higher Education, Research, Development and Innovation Funding (UEFISCDI) for financial support through the scientific research project no. 257.

REFERENCES

1. M. C. Daniel and D. Astruc, *Chem. Rev.,* **2004**, *104*, 293-346.
2. N. L. Rosi and C. A. Mirkin, *Chem. Rev*., **2005**, *105*, 1547-1562.
3. K. A. White and N. L. Rosi, *Nanomedicine*, **2008**, *3*, 543-553.
4. X. Huang, P. K. Jain, I. H. El-Sayed and M. A. El-Sayed, *Nanomedicine*, **2007**, *2*, 681-693.
5. O. Horovitz, G. Tomoaia, A. Mocanu, T. Yupsanis and M. Tomoaia-Cotisel, *Gold Bull*., **2007**, *40*, 213-218.
6. A. Mocanu, I. Cernica, G. Tomoaia, L. D. Bobos, O. Horovitz and M. Tomoaia-Cotisel, *Coll. Surf. A*, **2009**, *338*, 93-101.
7. G. Tomoaia, P. T. Frangopol, O. Horovitz, L. D. Boboş, A. Mocanu and M. Tomoaia-Cotisel, *J. Nanosci. Nanotechnol*. **2011**, *11*, 7762-7770.
8. R. D. Pasca, A. Mocanu, S. C. Cobzac, I. Petean, O. Horovitz and M. Tomoaia-Cotisel, *Particulate Sci, Technol*., **2014**, *32*, 131-137.
9. R. C. Fierascu, I. R. Bunghez, R. Somoghi, I. Fierascu and R. M. Ion, *Rev. Roum. Chim*., **2014**, *59*, 213-218.
10. C. P. Racz, S. Santa, M. Tomoaia-Cotisel, G. Borodi, I. Kacso, A. Pirnau and I. Bratu, *J. Incls. Phenomena and Macrocyclic Chem.,* **2013**, *76*, 193-199.
11. C. P. Racz, G. Borodi, M. M. Pop, I. Kacso, S. Santa and M. Tomoaia-Cotisel, *Acta Cryst.,* **2012**, *B68*, 164-170.
12. C. P. Racz, R. D. Pasca, S. Santa, I. Kacso, G. Tomoaia, A. Mocanu, O. Horovitz and M. Tomoaia-Cotisel, *Rev. Chim. (Bucharest),* **2011**, *62*, 992-997.
13. J. Szejtli, *Pure Appl. Chem*., **2004**, *76*, 1825-1845.
14. J. Szejtli, *Chem. Rev*., **1998**, *98*, 1743-1754.
15. E. M. M. Del Valle, *Process Biochem. (Oxford, U.K.)*, **2004**, *39*, 1033-1046.
16. Y. Liu, K. B. Male, P. Bouvrette and J. H. T. Luong, *Chem. Mater*., **2003**, *15*, 4172-4180.
17. P. Bouvrette, Y. Liu, J. H. T. Luong and K. B. Male, *US Patent*, 7232474 B2, 2007.
18. X. Y. Ling, I. Y. Phang, D. N. Reinhoudt, G. J.Vancso and J. Husken, *Int. J. Mol. Sci.,* **2008***, 9*, 486-497.
19. S. Jaiswal, B. Duffy, A. K. Jaiswal, N. Stobie and P. McHale, *Int. J. Antimicrob. Ag.*, **2010**, *36*, 280-283.
20. H. Li and Y.-W. Yang, *Chinese Chem. Lett.*, **2013,** *24*, 545-552.
21. A.V. Kabashin, M. Meunier, C. Kingston and J. H. T. Luong, *J. Phys. Chem. B*, **2003**, *107*, 4527-4531.
22. S. Pande, S. K. Ghosh, S. Praharaj, S. Panigrahi, S. Basu, S. Jana, A. Pal, T. Tsuduka and T. Pal, *J. Phys. Chem. C*, **2007**, *111*, 10806-10813.
23. T. Huang, F. Meng and L. Qi, *J. Phys. Chem. C*, **2009**, *113*, 13636-13642.
24. B. Aswathy, G.S. Avadhani, S. Suji and G. Sony, *Frontiers Mater. Sci.*, **2012**, *6*, 168-175.
25. X. Tian, C. Cheng, H. Yuan, J. Du, D. Xiao, S. Xie and M. M. F. Choi, *Talanta*, **2012**, *93*, 79-85.
26. R. J. Hunter, "Zeta potential in colloid science: Principles and applications", Academic Press, London, 1981.