Platinum Nanoparticles Prepared by Laser Ablation in Aqueous Solutions: Fabrication and Application to Laser Desorption Ionization

<table>
<thead>
<tr>
<th>Journal:</th>
<th>The Journal of Physical Chemistry</th>
</tr>
</thead>
<tbody>
<tr>
<td>Manuscript ID:</td>
<td>jp-2011-07324h.R1</td>
</tr>
<tr>
<td>Manuscript Type:</td>
<td>Article</td>
</tr>
<tr>
<td>Date Submitted by the Author:</td>
<td>n/a</td>
</tr>
<tr>
<td>Complete List of Authors:</td>
<td>Cueto, Maite; Consejo Superior de Investigaciones Científicas, Física Molecular Sanz, Mikel; Instituto Química Física Rocasolano Oujja, Mohamed; Instituto de Química Física Rocasolano, CSIC Gámez, Francisco; Universidad Pablo de Olavide Martinez-Haya, Bruno; Universidad Pablo de Olavide, Sistemas Físicos, Químicos y Naturales Castillejo, Marta; Instituto de Química Física Rocasolano, CSIC</td>
</tr>
</tbody>
</table>
Platinum Nanoparticles Prepared by Laser Ablation in Aqueous Solutions: Fabrication and Application to Laser Desorption Ionization

Maite Cueto¹,³, Mikel Sanz²*, Mohamed Oujja², Francisco Gámez¹, Bruno Martínez–Haya¹, Marta Castillejo²

¹Department of Physical, Chemical and Natural Systems, Universidad Pablo de Olavide, 41013 Seville, Spain

²Instituto de Química Física Rocasolano, CSIC, 28006 Madrid, Spain

³Instituto de Estructura de la Materia, CSIC, 28006 Madrid, Spain

*Email: mikel.sanz@iqfr.csic.es

RECEIVED DATE (to be automatically inserted after your manuscript is accepted if required according to the journal that you are submitting your paper to)
Abstract

Aqueous suspensions of platinum colloidal nanoparticles of varying size and polydispersity have been produced by ablation of a platinum target with a nanosecond Q-switched Nd:YAG laser. Several laser wavelengths (1064, 532 and 266 nm) and stabilizing agents (citrate and polymers PEG, PVA and PVP) were employed. Laser ablation with the infrared and visible wavelengths leads to spherical amorphous nanoparticles with a bimodal distribution of diameters, featuring a global maximum in the range 5-10 nm and a shoulder extending to 25 nm. Such bimodal distributions plausibly arise from thermal and explosive vaporization mechanisms occurring in different time scales, as proposed in earlier studies. Ultraviolet ablation of Pt at 266 nm, reported here for the first time, produces crystalline nanoparticles of small size (1-4 nm diameter), with a weak onset of larger particles (6-8 nm). The ablation at 266 nm also produced an appreciable yield of large rod-like particles of size ∼10 x 70 nm^2, especially in the presence of PEG and PVA. The Pt nanoparticles served to fabricate films capable of assisting the laser desorption ionization (LDI) of a model peptide. The best analytical performance for LDI mass spectrometric detection was obtained with Pt nanoparticles surface functionalized with citrate and PVA.
1. Introduction

Pulsed laser ablation has evolved as one of the most efficient physical methods for nanofabrication.\(^1\) In particular, pulsed laser ablation of solids in liquid environments (PLAL) has been extensively investigated as a technique for the fabrication of nanoparticles (NPs) of a large variety of materials including metals, alloys, ceramics, and semiconductors.\(^2-13\) Remarkable advantages of this method over chemical synthesis rely on the simplicity of the procedure, the weak agglomeration effects, and the lack of impurities caused by chemical precursors. The pioneering work on PLAL for materials processing was reported in 1987, \(^3\) where a metastable phase of iron oxide was synthesized by ablating an iron target in water. Since then, PLAL has been applied in the synthesis of nanoparticles of a range of materials, in particular metal nanoparticles of small dimensions and narrow size distributions.\(^3-13\) It has been shown that the nanoparticle concentration, size, polydispersity, shape and solid phase (amorphous or crystalline) can be controlled by the adequate choice of laser wavelength, pulse duration, and fluence (laser pulse energy per unit of the irradiated area).\(^12-17\)

Noble metal NPs have attracted much attention due to their unique plasmonic optical properties and potential applications as molecular sensors.\(^18-23\) More recently, they have contributed to the development of methods in mass spectrometry (MS), such as nanoparticle assisted laser desorption ionization (NALDI-MS).\(^24-27\) The NALDI methods benefit from the broadband optical absorption of noble metal NPs due to plasmonic resonances, which can range from the ultraviolet to the visible and near infrared.\(^27,28\) Among the noble metals, platinum is particularly advantageous for NALDI applications due to its low heat conductivity and high melting temperature, which favor the efficient local heating of the sample and lead to analyte desorption at lower laser powers. Yonezawa et al.\(^24\) have recently demonstrated the suitability of Pt NPs as NALDI substrates. However, platinum has been scarcely studied in NALDI-MS applications up to date; most studies have employed substrates made of gold and silver NPs.\(^27,29-34\)

Several works have shown that stable platinum colloidal nanoparticles can be prepared by PLAL in water solutions and that their properties depend on laser conditions (fluence and wavelength) as well as
on the stabilizing agent.\textsuperscript{35-40} Nichols et al.\textsuperscript{36} prepared platinum colloidal solutions at 1064, 532 and 355 nm using a nanosecond pulsed Q-switched Nd:YAG laser with high fluences (greater than 11 J cm\textsuperscript{-2}), well above the ablation threshold. It was found that the nanoparticle size distribution has a qualitatively similar bimodal shape at the three wavelengths, although a greater yield of large NPs was observed with increasing laser fluence and wavelength. Mafuné et al.\textsuperscript{38} and Park et al.\textsuperscript{40} observed that ablation at increasing surfactant concentration results in nanoparticles of smaller average diameter, because in this case the more efficient surface coating promotes faster completion of the growth process.\textsuperscript{38} On the other hand, Schwenke et al.\textsuperscript{17} have found that in stationary liquid, production of nanoparticles takes place not only by laser ablation of the target but also through fragmentation of dispersed nanoparticles by post irradiation.

In this work, platinum colloidal nanoparticles have been prepared by laser, ablating a platinum metallic target in water solutions of different polymeric stabilizing agents, using nanosecond pulses of 1064, 532 and 266 nm. A moderate range of fluences, just above the ablation threshold for each wavelength was selected in order to be able to control the mean diameter and size distribution of fabricated colloids. To our knowledge, production of Pt nanoparticles by PLAL at 266 nm had not been described previously. This study also addresses systematically the effect induced on the properties of the ablated NPs by stabilizing agents of different crystallinity and molecular weight. The results extend previous knowledge on the modulation of the size, polydispersity, shape and microscopic phase of the nanoparticles that can be achieved under different experimental conditions, namely laser wavelength, fluence and coating agent.

The optical properties of the colloidal solutions were investigated by UV-vis absorption spectroscopy, whereas the size, morphology and structure of the nanoparticles were characterized by transmission electron microscopy (TEM) and high resolution TEM (HRTEM). Finally, the efficiency as NALDI substrates of the different types of produced platinum colloids has been assessed by means of measurements with the model peptide Angiotensin I as analyte. It will be shown that ablation in citrate and PVA solutions yields Pt NPs most adequate for NALDI applications.
2. Experimental Section

2.1 Fabrication and characterization of platinum colloidal nanoparticles

A platinum target immersed in aqueous solution was ablated with the fundamental, second and fourth harmonics (1064, 532 and 266 nm) of a Q-switched Nd:YAG laser (Quantel, Brilliant B, 5 ns pulse duration) operating at a repetition rate of 10 Hz. The Pt metal target was placed at the bottom of a glass vessel filled with a 10 mm column (ca. 2.5 ml) of either pure water or a water solution of a stabilizing agent. The laser beam was focused by a 15 cm focal length lens situated approximately at 11-13 cm from the target to achieve a spot diameter of 1.2 mm for each wavelength. The diameter of the laser spot was measured from the trace left by the laser pulse on an uncoated PVC plate. Placing the laser focus after the Pt target has the advantage of reducing the potential interaction of the laser pulses with the ablation plume. The vessel was rotated at about 10 rpm during ablation to avoid target cratering. Laser fluences of 7.0, 3.5 and 1.5 J cm$^{-2}$, corresponding to 80, 40 and 17 mJ, lying close to the ablation threshold, were employed at the respective wavelengths of 1064, 532 and 266 nm. At these fluences, the luminosity of the ablation plume was of similar intensity at the three irradiation wavelengths. At 1064 nm, higher fluences of 10 and 14 J cm$^{-2}$ were also used to investigate their effect on the nanoparticle size distribution. For each set of parameters (laser wavelength, fluence and stabilizing agent), a nanoparticle dispersion was produced by irradiating the sample during 15 min (9000 pulses).

Experiments were conducted either in pure water or in aqueous solutions of four different stabilizing agents: Sodium Citrate (0.2 kDa, Panreac, 0.2 mM), Polyethylene Glycol (PEG, 12 kDa, Sigma-Aldrich, 0.01 mM), Polyvinyl alcohol (PVA, 10 kDa, Sigma-Aldrich, 0.01 mM) and Polyvinylpyrrolidone (PVP, 10 kDa, Sigma-Aldrich, 0.025 mM). PEG and PVA are typical crystalline polymers while PVP is amorphous. To investigate the effect of the molecular weight on the colloid properties, PEG with molecular weights of 1 kDa (Fluka, 0.07 mM), 6 kDa (Sigma-Aldrich, 0.01 mM) were employed. Deionized water, purified using a Mili-Q 18.2 MΩ reagent-grade water system from Millipore, was employed to prepare all the solutions.
The nanoparticle yields were characterized by measuring their UV-vis absorption spectra in a double beam spectrophotometer (Shimadzu UV-3600) using two 1 cm optical path quartz cuvettes for the sample and for the pure solvent as reference. For TEM and HRTEM characterization, the nanoparticle suspensions were spotted onto a carbon-coated Cu grid. TEM photographs were taken with a Philips CM-200 equipment and HRTEM images with a JEOL JEM-2100 system. Nanoparticle size analysis was performed using the image processing program ImageJ by measuring the diameter of 200-300 particles from 3 TEM images for each laser irradiation condition.

2.2 NALDI instrumentation and method

NALDI-MS measurements in positive-ion mode were performed in a commercial reflectron time of flight mass spectrometer (RETOF-MS, REFLEX III of Bruker Daltonics). The analyte ions were accelerated through a voltage drop of 20.0 kV, applied with a 0.25 µs delay with respect to the laser pulse. The reflectron was operated with a 24.7 kV voltage rise to reflect the ions toward the dynode-SEM (secondary electron multiplier) ion detector. Laser radiation of 337 nm was delivered by a N₂ laser (Bruker, NL110B) in the form of 4 ns pulses at a 4 Hz repetition rate. Laser pulses were applied with energies up to 20 µJ, focused on the sample with a lens of a 5 cm focal length, and mass spectra were averaged over 100 laser shots.

The analyte Angiotensin I (Sigma-Aldrich, 1296 Da) was prepared in a 0.15 mM working solution with a 7:3 of acetonitrile:water solvent mixture. The NALDI samples were prepared by mixing equal volumes of the working solution and the Pt nanoparticle suspension. Finally, a 2 µL sample aliquot was spotted and air-dried on the stainless-steel sample plate. For the NALDI threshold measurements, the laser pulse energy was slowly increased until an incipient analyte ion signal was observed in the mass spectrum, corresponding to Angiotensin I, either protonated or cationized by the Na⁺ or K⁺ ubiquitous impurities of the sample. In order to minimize “hot-spot” effects, the laser was continuously scanned through the sample during data collection. A series of NALDI mass spectra was collected at different pulse energies and the overall analyte ion yield was fitted linearly to obtain the threshold energy.
3. Results and Discussion

3.1 Characterization of the nanoparticles produced by laser ablation in solution

Figure 1 shows illustrative UV-vis spectra of the Pt NP suspensions produced in the present study. The spectra display broad absorption band structures decreasing in intensity from the UV towards the visible. According to the similar spectra recorded by Bigall et al.\textsuperscript{41} for monodispersed Pt NPs produced by chemical methods, the plasmonic band of Pt is located at around 250 nm in the small particle size limit (diameters smaller than 30 nm), and shifts towards the visible for large particles. The present NP suspensions are polydispersed with a negligible presence of particles with sizes larger than 30 nm (see below). Hence, the shoulder observed around 250 nm in the UV absorption spectra could be attributed to plasmonic excitation, possibly partially overlapping with interband transitions.\textsuperscript{36,38} We did not observe potential plasmonic bands better defined than those shown in Figure 1 for any of the samples. This is not surprising given the small size and appreciable polydispersity of the NPs produced by the ablation method, as discussed below. It was therefore not possible in our case to extract information on the platinum nanoparticle size from the recorded absorption spectra. As observed in Figure 1, the ratio of absorbances at 266 and 532 nm is approximately 50 for PVA, 20 for PVP and 5 for citrate, PEG and pure water.

The geometry and size distribution of the nanoparticles were determined by TEM. For illustration, Figure 2 displays typical TEM images of the NPs fabricated by laser ablation in pure water (i.e. without stabilizing agents) at the three laser wavelengths of this study. The size distributions of the nanoparticles extracted from each of the TEM images are also shown in Figure 2. All NP samples were found to display a bimodal distribution of sizes in which the large size component was enhanced at longer wavelengths and higher laser fluences. Accordingly, each distribution was deconvoluted into two components by best fits with a pair of lognormal functions. Table 1 provides the average sizes and widths of the two components, as well as their relative contribution to the global distribution, both in
terms of the number and of the mass of the NPs. Note that even a relatively small number density of the large NP component implies a substantial fraction of the mass of the ablated material.

Ablation in pure water led systematically to spherical nanoparticles with a low degree of agglomeration and a larger overall size at the longer ablation wavelengths. Figure 2a shows that at 1064 nm the global maximum of the size distribution is centered at 9 nm, whereas a broad secondary maximum is observed at about 19 nm. Increasing the fluence of the ablation laser up to 14 J cm\(^{-2}\) (Figure 2b) does not shift the distribution but the relative intensity of the large size component is enhanced. Ablation at 532 nm in pure water (Figure 2c) also produced NPs with a relatively broad size distribution, extending above 20 nm. The bimodal shape is in this case less marked than that found for ablation at 1064 nm, and the distribution could be fitted with peaks centered at 7 nm and 14 nm. Finally, ablation at 266 nm (Figure 2d) leads to significantly smaller nanoparticles with sizes confined within 1-4 nm, complemented by a weak onset of a bimodal component made of larger NPs with diameters in the 6-8 nm range that is clearly visible in the TEM image.

The NP size distributions described above are in consonance with previous studies of laser ablation of platinum in water using the wavelengths of 1064, 532 and 355 nm, although the laser fluences (11-65 J cm\(^{-2}\)) used were significantly greater than those applied here.\(^{36}\) In those studies, similarly broad and bimodal NP size distributions were observed, also narrowing down and shifting to smaller sizes with decreasing wavelength. The two components of the NP size distribution were plausibly traced back to a thermal vaporization mechanism (small NP component) and to an explosive ablation mechanism (large NP component) taking place in different time scales.\(^{36}\) These mechanisms presumably also apply in our experiments and are briefly discussed below in Sect. 3.2.

We consider now the effect of the stabilizing agent on the properties of the nanoparticles. The addition of the polymeric surfactants PEG (12 kDa), PVA (10 kDa) and PVP (10 kDa) had a relatively small effect on the size of the NPs. Table 1 shows that the nanoparticles produced in solutions of these surfactants display size distributions that are comparable to those obtained in pure water, perhaps with the exception of the ablation at 532 nm where a significant NP size reduction is observed. On the other
hand, the use of citrate as stabilizing agent reduced appreciably the size of the ablated NPs in a systematic way. Figure 3 shows TEM images and size distributions of the Pt NPs produced in citrate and PVP solutions, and illustrates the considerable smaller nanoparticle sizes and lower polydispersity achieved in citrate in comparison to ablation in pure water or in the polymeric solutions. Motivated by this observation, the ablation experiments were repeated with PEG polymers of smaller molecular weight, namely 1 and 6 kDa. However, no significant changes were observed in the properties of the NPs synthesized with these smaller molecular weight PEGs at any of the irradiation wavelength. It is worth noting that the stabilization of the citrate anionic coating is driven by stronger electrostatic interaction than that of the neutral polymeric agents. Hence, the smaller NP size is likely to be due to the efficient coating of the nascent positively charged nanoparticles during the ablation process, favored by the negative charge of citrate molecules in the dissolved state.

HRTEM images of the deposited solutions were recorded in order to gain insight into the crystalline properties of the nanoparticles. Upon infrared (1064 nm, Figure 4a) and visible (532 nm, not shown) ablation, signs of crystallinity were not evident. On the other hand, ablation by UV irradiation (266 nm) led to platinum nanoparticles displaying the typical features of a Pt crystalline lattice (Figures 4b,c). The measured lattice interplane distance is 0.23 nm (Figure 4b), which matches well the corresponding value for the (111) orientation of cubic, single crystalline platinum. The Fourier transform of the nanoparticle image (in the inset of Figure 4b) corroborates the crystalline character of the NPs obtained at this short wavelength.

Interestingly, ablation at 266 nm was also differentiated from ablation at 532 nm and 1064 nm by the observation of an appreciable yield of larger anisotropic nanoparticles, in addition to the bulk of crystalline spherical NPs described above. Crystalline platinum rod-like particles with dimensions around 10 x 70 nm$^2$ were obtained in the suspensions. Figure 4c depicts HRTEM images of such nanorods showing that the crystal interplane distance is again around 0.23 nm, as found for the spherical nanoparticles. Several repetitions of the ablation synthesis procedure led to qualitatively similar results. Whereas the crystalline nanorods could be observed in all the samples produced by ablation at 266 nm,
their presence was appreciably enhanced when the crystalline polymers PEG and PVA were employed as stabilizing agents. The reason for these findings is unclear, and some speculative mechanisms are outlined in Section 3.2.

3.2 Ablation process and nanoparticle growth mechanisms

In the present work, Pt NPs have been produced by laser irradiation of a Pt metal target at fluences just above ablation thresholds, namely 7.0, 3.5 and 1.5 J cm\(^{-2}\) at 1064, 532 and 266 nm, respectively. These fluence values yielded similar plume luminosities and are consistent with the decrease in the reflectivity (i.e. increase of absorptivity) at shorter wavelengths, measured for Pt in this spectral range (reflectivities of ca. 0.85, 0.70 and 0.50 at 1064, 532 and 266 nm, respectively).\(^{42-44}\) The non-reflected laser radiation penetrates into the Pt target a distance (optical depth) inversely proportional to the absorption coefficient. The optical depths of Pt are of ca. 12, 11 and 9 nm at 1064, 532 and 266 nm, respectively.\(^{43,45}\) These values anticipate decreasing ablation rates as the laser wavelength becomes shorter,\(^{46}\) leading to a smaller absolute amount of material ablated from the sample and participating in the NP formation and growth mechanisms. It is also expected that irradiation of the platinum target at shorter wavelengths leads to a more uniform target ablation, and to a more vaporized plasma, mainly formed by small clusters and a large percentage of atomic species. Irradiation at longer wavelengths will in turn favor explosive ejection of molten droplets.\(^{35-37}\) These features aid in explaining the general observation that the ablated nanoparticles have gradually smaller overall size as the laser wavelength becomes shorter.

In addition to the direct penetration of the laser into the material, thermal propagation occurs during the interaction of the nanosecond laser pulse with the solid target. The thermal wave propagates from the irradiated volume into the sample at a rate determined by the thermal diffusivity of the material, \(D = 0.25 \text{ cm}^2 \text{ s}^{-1}\) for Pt,\(^{47}\) which leads to a heat diffusion length of ca. \(l_{th} = 0.7 \mu \text{m}\) during the 5 ns pulse duration of the ablating laser. Local heating of the Pt target material is then given by \(\Delta T = H (\rho C_P l_{th})^{-1}\), where \(\Delta T\) is the increase of temperature, \(\rho\) and \(C_P\) are the density and thermal capacity of the material.
(21.5 g cm\(^{-3}\) and 25.9 J mol\(^{-1}\) K\(^{-1}\) for Pt, respectively) and \(H\) is the energy absorbed per unit area in each laser pulse. Considering the laser fluences employed in our study, corrected from the reflectivity values given above at each wavelength, local target temperatures ranging from 4000 K (at 266 nm) to 5000 K (at 1064 nm) are estimated.

The bimodal character of the size distribution of the NPs prepared by laser ablation of a metallic target immersed in water has been related in previous works to the participation of two qualitatively different NP formation mechanisms.\(^{13,35-38}\) The small NP size component is attributed to vaporization of atoms and small clusters from the irradiated target surface layer,\(^{36}\) seeding further growth in the ablation plume.\(^{38}\) Immediately after ablation, a dense cloud of atoms and clusters builds up above the laser spot on the target. Particle growth by sequential aggregation follows, as long as the pair interactions between the atoms and clusters are stronger than their interactions with the solvent and surfactant molecules of the aqueous phase. The aggregation process continues rapidly in the plume until atoms in the close vicinity are almost completely consumed. Within this nucleation and growth mechanism the size distribution becomes pure lognormal,\(^{38}\) in good agreement with our observations for the small size component of the NP distribution observed at the three wavelengths investigated.

On the other hand, the large size component of the NP size distribution is ascribed to nanoparticles resulting from explosive boiling of the target, leading to the ejection of molten material. Nucleation through random thermal fluctuations of small amount of vapor inside the irradiated target promotes bubble growth. Bubbles move toward the surface of the target where they explosively break through, ejecting a mixture of droplets and vapor.\(^{8}\) This pathway can be expected to be enhanced by the greater penetration depth of the longer laser wavelengths, which would explain the greater yield of large particles and the more marked bimodal character of the NP size distribution in the ablation at 1064 nm, in comparison to 532 and 266 nm.

It must be remarked that the interactions and dynamics of the ejected material within the ablation plasma are actually quite complex and involve a different balance of fundamental processes at each irradiation wavelength. For instance, absorption of laser photons by the plasma through inverse
Bremsstrahlung\textsuperscript{48} is, on one hand, considerably more favorable for IR than for UV wavelengths. On the other hand, photoionization in the ablation plasma is more efficient for the UV. Furthermore, the varying collision probability due to plasma heating of the ablated species with the liquid environment can contribute to the broad range of size distributions observed experimentally.\textsuperscript{49}

Direct laser fragmentation of the plume material and of the nanoparticles suspended in the solution induced by the laser pulses has been postulated to explain the small nanoparticle sizes produced by PLAL of silver targets.\textsuperscript{17,50} Nevertheless, in the case of ns PLAL of platinum targets in stationary regime at wavelengths in the IR-UV range, this mechanism was considered not to contribute appreciably to nanoparticle production,\textsuperscript{36,38} even at fluences significantly higher than those applied here. As shown in reference\textsuperscript{36}, irradiation at 1064 nm, where the absorption coefficient of platinum NPs is negligible, leads to a small size component, attributed to a thermal vaporization mechanism, with similar mean size and width to those obtained under 532 and 355 nm ablation, two wavelengths where the absorption coefficient is higher. Photofragmentation effects of the plume material were also found to be negligible for ablation of other metals, such as Zn and Mg, in liquid solutions.\textsuperscript{17} In the present case it has been observed that the reduction of the mean NP diameter when the wavelength decreases from 532 to 266 nm, is similar for the stabilizing agents citrate and PVP (Table 1, Figure 3), even if the absorbance increases by a factor of 5 for citrate and 20 for PVP (Figure 1) in this wavelength range.

The use of stabilizing agents is expectedly found to reduce the size of the nanoparticles. In pure water solutions, the supply of platinum atoms outside the thermal nucleation region through diffusion causes the particles to keep growing at a slower pace, even after the rapid growth ceases. In the presence of a stabilizing agent, the slow growth stage tends to be completed by the coating of the particle surface. At each of the three irradiation wavelengths, the smallest nanoparticles were systematically obtained in citrate solutions due to the more efficient coating of the ablated material. On one hand, the coating is stabilized by electrostatic interaction and, on the other hand, the diffusion coefficient of citrate in the solution is larger than those of the polymeric surfactant molecules due to its much smaller size.\textsuperscript{38} As already mentioned, ablation in the presence of PEG with a molecular weight as low as 1 kDa (but a
factor of about five greater than that of citrate) still yielded NPs much larger in size than those obtained with citrate as stabilizing agent.

One remarkable observation of our study has been the predominance of crystalline nanoparticles by ablation at 266 nm, in contrast to the amorphous character of the nanoparticles produced at 532 and 1064 nm. This result was not necessarily expected, as this ablation wavelength had not been explored in the past, and no reference to crystalline particles was reported in previous ablation studies in the near UV, namely at 355 nm. Crystallinity plausibly arises from the rapid ejection of relatively cool material from the outermost layers of the target, which can also be related to the small particle sizes observed at 266 nm. In addition, crystalline platinum nanoparticles can be formed by molecular diffusion (Ostwald ripening) and coagulation followed by coalescence of the platinum atoms and small clusters present in the vaporized plume. Any substantial contribution from the explosive boiling channel then seems to be ruled out in the ablation induced at this short wavelength.

In this scenario, it has been striking to find a systematic production of large crystalline nanorods in the ablation at 266 nm. The production of nanorods was particularly enhanced in the presence of the two crystalline polymers, PEG and PVA. These two polymers are more likely than citrate and PVP to build up crystalline micellar cores on the surface of the nanoparticles. PEG and PVA coat the Pt particles through weak electrostatic interactions while exposing the more hydrophilic groups outward to the aqueous phase. As shown, molecular weight of the PEG stabilizing agent (1, 6 and 12 kDa) did not affect the size of nanorods while their density decreased at increasing molecular weight. The formation of nanorods plausibly arises from coalescence of the ablated material, with the surfactants acting not simply as templates but also as shape-inducing reagents, in a qualitatively similar way as found in the case of PLAL of Mg in a previous study. Pyrolysis of the polymeric coating by the 266 nm laser radiation may as well aid in the formation of the nanorod particles. However the relevance of such potential mechanism cannot be confirmed at the present stage. On the other hand, post-ablational irradiation effects, as discussed earlier in relation to the production of small spherical NPs at 266 nm, can also be considered as influencing the generation of nanorods. However, at the laser fluences
employed here, irradiation of the ablated material dispersed in solution should be a process of minor relevance. In fact, the stability of the large nanorods at 266 nm can be taken as a probe of the limited action of the laser pulses on the dispersed material.

3.3 Nanoparticle assisted laser desorption/ionization.

A systematic NALDI investigation was carried out by preparing active substrates with a selection of platinum nanoparticle samples produced by laser ablation. The peptide Angiotensin I (henceforth, Ang) was chosen as model analyte due to its relatively low molecular weight and representative aminoacid sequence. Angiotensin I is particularly advantageous for NALDI studies because the nitrogen sites of its His and Arg residues are easily cationized by the ubiquitous Na$^+$ and K$^+$ impurities of the sample.$^{53}$ In this way, the need for efficient proton sources to ionize the analyte is avoided.

It was rewarding to find that all the Pt NP samples had a reasonable performance when employed as NALDI substrates. This was not necessarily expected in view of the appreciable polydispersity that characterized the nanoparticles produced by laser ablation. Figure 5 shows three illustrative NALDI-MS spectra of Angiotensin I recorded on a substrate of produced with the platinum NPs fabricated by laser ablation at 532 nm in citrate (Figure 5a), in PVA solution (Figure 5b) and in pure water (Figure 5c) as reference. The more intense peaks are assigned to the protonated adduct [Ang + H]$^+$, and to the association with one or more alkali cations, [Ang + Na]$^+$, [Ang + K]$^+$ and [Ang + 2Na - H]$^+$, where Ang stands for Angiotensin I. Interestingly, the use of substrates prepared with the NPs ablated in water, citrate or PEG led systematically to a predominance of the [Ang+Na]$^+$ adduct signal (Figures 5a and 5c), whereas the PVA and PVP surfactants proved to be more efficient proton donors and yielded preferentially protonated Angiotensin adducts (Figure 5b). These results indicate that through careful control of the stabilizing agent it is possible to achieve selective analyte ionization for NALDI applications. A more systematic study is in progress to draw definite conclusions to this respect.

The NALDI efficiency of the NP substrates was evaluated in terms of the laser pulse energy threshold required for analyte detection, taking as reference the most intense adduct, [Ang+H]$^+$ or [Ang+Na]$^+$. 
Whereas NALDI-MS spectra of fair quality could be recorded with substrates of all the nanoparticle samples, differences in performance were observed leading to detection thresholds in the range 2-12 µJ. No analyte signal was detected in test NALDI samples prepared from solutions of the surfactants and Angiotensin without nanoparticles. The lowest thresholds for NALDI detection (ca. 2-4 µJ) were obtained with the Pt NPs fabricated by ablation at 532 nm, in particular with those coated with citrate (for [Ang+Na]^+ detection) and with PVA (for [Ang+H]^+ detection) as stabilizing agents. The NPs produced at 1064 nm yielded a comparably smaller NALDI efficiency, which can be attributed to their appreciably higher polydispersity. In this sense, the NPs ablated at 532 nm seem to have a balance of size and polydispersity appropriate for NALDI. These nanoparticles are relatively small, especially the ones produced with the citrate coating (see Figure 3), but they still have a significant absorptivity at the wavelength of 337 nm of the laser employed in the NALDI experiments. The small heat capacity of the NPs due to their small size can also be expected to induce a rapid heating of the substrate, leading to efficient analyte desorption. The citrate coating provides a more homogeneous and closer interaction between the nanoparticle and the peptide analyte (Angiotensin I) than the bulky polymeric surfactants PEG, PVA and PVP. The polymers act as molecular spacers separating nanoparticle and analyte and thus tend to require higher laser energies to efficiently transfer sufficient heat to the analyte to assist desorption. In this sense, the good NALDI performance of the PVA coated NPs was somewhat surprising and must be attributed to a favorable pathway for the protonation of the analyte. In fact, the laser energy threshold for detection of the sodiated adduct was in this case at least a factor of three higher than the one measured for the citrate coated nanoparticles. It is also worth noting that the bare nanoparticles produced by ablation in pure water constituted comparably poor NALDI substrates and yielded spectra with low signal to noise ratios (see Figure 5c). This finding can be related to the agglomeration of the nanoparticles as the solvent is allowed to evaporate during the NALDI sample preparation process in the absence of surfactants, thereby reducing the interfacial area available for interaction with the analytes. The use of appropriate coating agents seems then of key importance for NALDI applications.
4. Conclusions

Nanosecond laser ablation at three wavelengths of the Nd:YAG laser (1064, 532 and 266 nm) has been used to produce platinum nanoparticles in pure water and in various aqueous solutions of stabilizing agents (citrate, PEG, PVA, PVP). The results show that the nanoparticle size distribution can be controlled with the appropriate choice of both laser wavelength and stabilizing agent. Citrate demonstrated a greater ability than the polymeric surfactants to abort the nanoparticle growth, thereby reducing the final size of the ablated colloids. This finding is attributed to the effect of electrostatic stabilization produced by the interaction between the negatively charged citrate molecules in the dissolved state and the positively charged nanoparticles and to the greater diffusion coefficient of citrate into the ablation plume.

The distribution of sizes of the ablated nanoparticles is bimodal, with a small size component presumably associated to a thermal ablation and growth mechanism, and a large size component expected to correlate with an explosive ablation mechanism. This latter pathway is enhanced at the longer ablation wavelengths (532 and 1064 nm), which also leads to an overall increase in the average size and polydispersity of the nanoparticles. In contrast, ablation in the ultraviolet (266 nm) produces predominantly NPs of small size (< 5 nm). Short wavelength ablation also yields crystalline nanoparticles, in contrast to the amorphous structure achieved at the longer wavelengths. These features have been ascribed to the production of a more vaporized plasma (mainly formed by small clusters and a large percentage of atomic species) at this shorter wavelength and in a minor extent to the fragmentation of dispersed nanoparticles by post irradiation. In addition, ablation at 266 nm produces an appreciable yield of large crystalline nanorods, especially when the crystalline stabilizing agents, PEG and PVA, are employed.

The platinum nanoparticles produced by laser ablation have demonstrated a fair capability for assisting laser desorption/ionization mass spectroscopy detection of a model peptide (Angiotensin I). The study has also corroborated that the coating agent also serves to modulate the ionization
mechanism: whereas citrate or PEG lead systematically to the predominance of the analyte cationized by the Na\(^+\) impurities, the PVP and PVA surfactants yielded preferentially protonated adducts. The lowest NALDI thresholds and best quality mass spectra were obtained with nanoparticles fabricated in aqueous citrate and PVA solutions for the detection of sodiated and protonated adducts, respectively. This result is plausibly due to the favorable balance between size and polydispersity of the nanoparticles, to the tight interactions between the nanoparticles and the peptide analyte induced by the low molecular weight of the citrate coating agent, and to the efficient protonation of the peptide by the PVA polymer during the NALDI process.

Acknowledgments

Funding from Ministry of Science and Innovation of Spain (MICINN) under Projects CTQ2009-10477, CTQ2010-15680, CONSOLIDER-INGENIO CSD2007-00058 and CSD2009-00038, and Programa Geomateriales (CAM, S2009/Mat-1629) are gratefully acknowledged. Funding from the Operative Programme FEDER-Andalucia 2007-2103 through projects P07-FQM-02600 and P09-FQM-4938 also contributed to the present research. We thank GEMPPO at the IEM-CSIC for the use of the HRTEM and CITIUS from Universidad de Sevilla for technical assistance and the use of their TEM equipment.
References


[48] Laser-Induced Breakdown Spectroscopy; Russo, R. E.; Mao, X. L.; Yoo, J. H.; Gonzalez, J. J.


Table 1. Average diameter D, width W (full width at half maximum) and relative mass contribution M of the two components of the bimodal size distribution of Pt nanoparticles fabricated by irradiation at the indicated wavelengths, in pure water and aqueous surfactant solutions. Laser fluences of 7, 3.5 and 1.5 J cm$^{-2}$, lying close to the ablation threshold, were employed at the respective wavelengths of 1064, 532 and 266 nm. At 266 nm the main component could only be evaluated in pure water and in PVP, which showed an ensemble of larger particles of sufficient statistical relevance. The large nanorods observed by ablation at 266 nm are excluded from the analysis.

<table>
<thead>
<tr>
<th>Stabilizing agent</th>
<th>(\lambda) (nm)</th>
<th>Small size component</th>
<th>Large size component</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>D (nm)</td>
<td>W (nm)</td>
</tr>
<tr>
<td>none</td>
<td>1064 nm$^a$</td>
<td>9</td>
<td>7</td>
</tr>
<tr>
<td></td>
<td>1064 nm</td>
<td>9</td>
<td>7</td>
</tr>
<tr>
<td></td>
<td>532 nm</td>
<td>7</td>
<td>6</td>
</tr>
<tr>
<td></td>
<td>266 nm</td>
<td>3</td>
<td>2</td>
</tr>
<tr>
<td>PEG</td>
<td>1064 nm</td>
<td>11</td>
<td>7</td>
</tr>
<tr>
<td></td>
<td>532 nm</td>
<td>4</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td>266 nm</td>
<td>3</td>
<td>2</td>
</tr>
<tr>
<td>PVA</td>
<td>1064 nm</td>
<td>9</td>
<td>6</td>
</tr>
<tr>
<td></td>
<td>532 nm</td>
<td>4</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td>266 nm</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>PVP</td>
<td>1064 nm</td>
<td>10</td>
<td>6</td>
</tr>
<tr>
<td></td>
<td>532 nm</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td></td>
<td>266 nm</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>Citrate</td>
<td>1064 nm</td>
<td>6</td>
<td>5</td>
</tr>
<tr>
<td></td>
<td>532 nm</td>
<td>4</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td>266 nm</td>
<td>2</td>
<td>1</td>
</tr>
</tbody>
</table>

$^a$ fluence 14 J cm$^{-2}$
Figure Captions

Figure 1. Optical absorption spectra of the solutions obtained upon 266 nm laser irradiation in deionized water and in aqueous solutions of different stabilizing agents as indicated.

Figure 2. TEM images of nanoparticles fabricated in pure water by ablation of a platinum target at: a) 1064 nm, 7.0 J cm\(^{-2}\), b) 1064 nm, 14.0 J cm\(^{-2}\), c) 532 nm, 3.5 J cm\(^{-2}\), and d) 266 nm, 1.5 J cm\(^{-2}\). The right column presents the corresponding nanoparticle size distribution. Solid lines correspond to fittings (see text).

Figure 3. TEM images of nanoparticles fabricated by laser ablation of platinum at: a) 532 nm, 3.5 J cm\(^{-2}\), and b) 266 nm, 1.5 J cm\(^{-2}\) in aqueous solutions of citrate and at: c) 532 nm, 3.5 J cm\(^{-2}\), and d) 266 nm, 1.5 J cm\(^{-2}\) in aqueous solutions of PVP. The right column presents the corresponding nanoparticle size distributions. Solid lines correspond to fittings (see text).

Figure 4. HRTEM images of the typical spherical nanoparticles fabricated by laser ablation of platinum in water at: a) 1064 nm and b) 266 nm. The upper-right inset in b) shows the corresponding Fourier transform. c) Images of typical nanorods (left) and magnification (right) showing their crystalline structure. The nanorods were particularly abundant in the suspensions produced by 266 nm ablation in PEG and PVA solutions.
Figure 5. NALDI RETOF-MS of Angiotensin I (Ang) recorded by using a substrate of stained steel with deposited platinum colloidal nanoparticles fabricated by laser ablation at 532 nm in aqueous solution of:

a) citrate, b) PVA and c) pure water.
Figure 1

- PVA, 10 kDa
- PVP, 10 kDa
- Citrate
- PEG, 12 kDa
- Water

Absorbance vs. Wavelength (nm)

Key wavelengths:
- 266 nm
- 532 nm
Figure 2

a) 1064 nm, 7 J/cm²

b) 1064 nm, 14 J/cm²

c) 532 nm

d) 266 nm
Figure 3

(a) Citrate, 532 nm

(b) Citrate, 266 nm

(c) PVP, 532 nm

(d) PVP, 266 nm
Figure 4

a) Water, 1064 nm

b) Water, 266 nm

0.23 nm

5 nm

10 nm

c) PVA, 266 nm
Figure 5

a) Citrate

[bbox:0.35 0.47 0.65 0.5]

b) PVA

[cbox:0.35 0.6 0.65 0.67]

c) Water

[dbox:0.35 0.75 0.65 0.83]