

# Assessment of Biomass of Leaves of Water Hyacinth (*Eichhornia crassipes*) as Reducing Agents for the Synthesis of Nanoparticles of Gold and Silver

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**Abstract**— Green chemistry methods for nanoparticles synthesis have implemented the valorization of renewable waste that reduces the use of chemicals and sub-products to minimize the environmental impact. Herein, we report a method to synthesize Ag and Au nanoparticles (AgNPs, AuNPs) using one of the world's worst aquatic weeds, water hyacinth. From a reaction between a solution of AgNO<sub>3</sub> or HAuCl<sub>4</sub> and controlling the pH, the nanoparticles were synthesized. The optimum pH value to obtain uniform quantum dots was found to be acidic for AgNPs and neutral for AuNPs. The size was highly dependent on pH for AgNPs, a smaller size was for acidic pH, and the larger size was for basic pH, and cubic and hexagonal are the predominant structures, no dependent was observed in AuNPs, and orthorhombic is the most common form. This method was sustainable because water hyacinth is a renewable resource in all world, and their use is not being exploited in any process. The bioreduction process using water hyacinth promotes the metallic nanoparticles formation and applied standard conditions for temperature and pressure. Also, the rate of synthesis is fast.

**Keywords**— Assessment of water hyacinth, green synthesis, gold nanoparticles, leaf biomass, silver nanoparticles.

## I. INTRODUCTION

The study of the metal bioaccumulation/phytoremediation process in plants has revealed that metals are deposited as nanoparticles in various organs and tissues (Makarov et al., 2014). A nanoparticle is defined as a discrete particle that has a structure in the nanometer size range, usually from 1 to 100 nm. The high surface area due to volume ratio of nanoparticles makes their physicochemical properties quite different from those of the bulk material

(Hebeish, El-Rafie, El-Sheikh & El-Naggar, 2013; Ahmad, 2014). Many researchers have been focused on silver and gold nanoparticles (AgNPs, AuNPs) in various fields of applications. Metal ion adsorption Bahadar et al., 2014; antimicrobial activity Muhammad et al., 2014; photocatalytic activity Wei et al., 2014; chemical sensors Rahman, Khan, Jamal, Faisal & Asiri, 2012 and, labeling Urusov et al., 2015.

Generally, AgNPs and AuNPs are synthesized by several methods whether physical or chemical. Thermal decomposition Hosseinpour & Ramezani, 2014; sonochemical Mohd & Ashokkumar, 2015; Darroudi, Zak, Muhamad, Huang & Hakimi, 2011; solvothermal Choi et al., 2013; microemulsion Ahmad, Wani, Al-Hartomy, Al-Shihri & Kalam, 2015; Jurkin, Guliš, Dražić, & Gotić, 2016; and laser ablation Urusov et al., 2015. The most of these methods need controlled environments and involve the use of hazardous chemical reagents like sodium borohydride and hydrazine hydrate (Hebeish et al., 2013). The request for an environmentally sustainable synthesis method has led to green chemistry techniques (Shameli et al., 2012). This approach could have a reduction/elimination of toxic and hazardous substances in the synthesis processes of metallic nanoparticles. Green chemistry uses microorganisms, enzymes, algae, plants (Majeed et al., 2016) and biomolecules (Hebeish et al., 2013).

The water hyacinth plants were considered as a good source in metal nanoparticles synthesis; it is contemplated as a weed because of growth in rivers and streams. It has been recognized as a hyperaccumulator organism in which high concentration of tannins makes it an excellent tool for heavy metals removal and metal nanoparticles synthesis (Rosano-Ortega et al., 2007).

Different parts of plants, such as leaves roots, stems, fruits, seeds, flowers, and latex; have been used for AgNPs and AuNPs synthesis (Rashidipour & Heydari, 2014; Mohd et al., 2015; Wang et al., 2016; Shameli et al., 2012; Babu, Aishwarya, Vidya & Saidutta, 2014; Heydari & Rashidipour, 2015; Baghizadeh, et al., 2015; Baharara, Namvar, Ramezani, Hosseini, & Mohamad, 2014; Ankamwar, Gharge & Sur, 2015; Guidelli, Ramos, Zaniquelli & Baffa, 2011); Various phytochemical compounds, including terpenoids, polyphenols, sugars, alkaloids, phenolic acids, and proteins, play a major role in the bioreduction of metals (Makarov et al., 2014). Tannic acid, a polyphenolic compound, has been used as a reductant agent, in which case phenols take part in the redox reaction by forming Quinones and donating electrons that reduce oxidized metal ions to form nanoparticles (Ahmad, 2014).

Rosano-Ortega et al. (2006) proposed an application of the water hyacinth biomass for the Mn reduction from a MnSO<sub>4</sub> solution and observed the dependence of the nanoparticle's size as follows pH conditions, in which the smallest clusters (1-4 nm) were generated using a pH=5. Thus a change in pH gave it a modification of the chemical state (ionization) of the phytochemicals in plants, which affects their capacity to reduce metal ions (Makarov et al., 2014; Rashidipour et al., 2014).

Water hyacinth was considered as a research subject due to the following characteristics:

- i). It is an aquatic weed, shared and available all over the world.
- ii). Accepts polyphenolic compounds in all sections of plants; But leaves contain a greater amount of these due to the plant's defense mechanism (Rashidipour et al., 2014). Because of these characteristics, polyphenolic compounds contribute to the reduction of silver and gold ions.
- iii). Also, the leaves take less time than the roots and stem in the reduction process.

For this study, a sustainable method was proposed for AgNPs and AuNPs synthesis using water hyacinth's leaves. Also, the pH effect was evaluated to optimize the synthesis route; in the same way, composition, size, and structure of the AgNPs and AuNPs were performed by using high-resolution electron microscopy.

## II. MATERIALS

Hydro chloroauric acid trihydrate (HAuCl<sub>4</sub> 3H<sub>2</sub>O, 99.9%) and silver nitrate (AgNO<sub>3</sub>, 99.0%) was purchased from Sigma-Aldrich, buffer solutions for pH 5 and seven were obtained from MERCK and for pH 9 and 11 from HYCEL. The water hyacinth was collected from Valsequillo Reservoir, Puebla, State in Mexico. All aqueous solutions were made using try distilled water.

## III. METHOD

**Biomass process:** Water hyacinth leaves were washed in running tap water, and received a second washing in try distilled water until it was evident. Then, the leaves were cut into small pieces and dried by solar radiation for 3h. The pieces were pulverized with a mortar and pestle and then with a blender. The obtained powder was washed with a solution of HCl 0.01N because it was required to break the cellulose, which improves the yield of sugars and release of phytochemicals, and finally it was dried again in a preheated oven at 80°C for 24h. The final powder was considered the biomass and was selected through a sieve (mesh #80) of diameter  $\leq 0.098$ in.

**Synthesis of nanoparticles:** Silver and gold nanoparticles were obtained according to the previously described method with slight modifications (Rosano et al., 2006; Schabes et al., 2006; Canizal et al., 2006); the flow of the process is shown in Figure 1. A homogeneous suspension of biomass was prepared with a concentration of 5 mg/mL in try distilled water using an ultrasonic bath (15 min). The pH of the solution was controlled by a buffer solution to the values of 5, 7 and 9 for AgNPs and 5, 7 and 11 for AuNPs. After placing the solutions in an ultrasonic bath (15min), it was centrifuged (3000 rpm) for 30 min. Then 25 mL of a silver solution  $3 \times 10^{-4}$  M of AgNO<sub>3</sub> or gold solution  $3 \times 10^{-4}$  M of HAuCl<sub>4</sub> was added. The samples were mixed homogeneously using an ultrasonic bath (15 min) and centrifuged (3000rpm) for one h. Finally, the biomass was separated from the solution by vacuum filtration using a filter with pore size of 0.45  $\mu$ m. The final settlement was kept at rest for 48 h. The process was carried out at steady conditions.

**Characterization of nanoparticles:** The analysis by TEM and HRTEM was carried out, using a JEOL JEM-2010F microscopy with a FasTEM for 2.3Å resolution. A couple of drops of the AgNPs or AuNPs solutions were deposited on a copper grid covered with amorphous carbon. HRTEM were obtained at various defocus conditions, including the optimal (Scherzer condition). Finally, Fast Fourier Transforms (FTT) was obtained from the images to identify the crystalline structure of each particle.

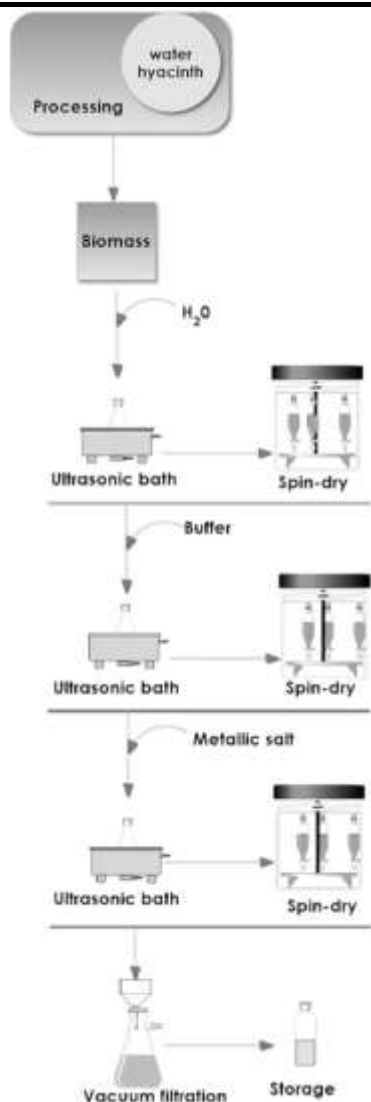


Fig.1: Schematic process of silver and gold nanoparticles synthesis with the biomass of leaves from water hyacinth.

Table captions appear centered above the table in upper and lower case letters. When referring to a table in the text, no abbreviation is used and "Table" is capitalized.

#### IV. RESULTS AND DISCUSSION

Particle Size Distribution (PSD). Different samples have been achieved by varying the pH value. The phytochemicals achieved with a variety of pH allowed to obtained AgNPs and AuNPs with a small size distribution and different configurations. TEM measurements were carried out to determine the size of AgNPs and AuNPs, also for both of them, in the micrographs, it was seen a trimodal size distribution (small, medium and large). Figure 2 shows the PSD for AgNPs. Here it can be observed changes in particle size due to pH value. In Figure 2a for pH= 5, a large number of nanoparticles were in 1-10 nm, for pH= 7 a great mix of particles were detected for 1-40 nm (Figure 2b) and at pH=9 the

majority of nanoparticles are in 1-20 nm (Figure 2c). To confirm this behavior; in Table 1 was included the finished size for nanoparticles,  $9.3 \pm 7.1$  nm,  $21.93 \pm 14.46$  nm and  $26.3 \pm 34.66$  nm for pH 5, 7 and 9 respectively. As follows these results at acidic pH small-sized nanoparticles were formed, whereas at neutral pH a large number of nanoparticles have been trained.

Table.1: Size, structure and composition of Ag and Au nanoparticles obtained with leave's biomass of water hyacinth.

Met al	Precur sor	pH	Avera ge Size	Comp osition	Perce nt	Struct ure	Plane
Ag	AgNO <sub>3</sub>	5	9.3±7.1	Ag <sup>0</sup>	86%	FCC	[111], [102]
		7	21.93 ± 14.46	UD	UD	UD	UD
		9	26.3± 34.66	Ag <sub>6</sub> O <sub>2</sub>	80%	HP	[001]
Au	HAuCl <sub>4</sub> *3H <sub>2</sub> O	5	6.3± 3.3	Au <sup>0</sup>	81%	UD	[111]
		7	4.2±1.79	AuO	68%	UD	UD
		11	47.8± 38	Au <sub>2</sub> O <sub>3</sub>	90%	UD	[511]

FCC: Face-centered cubic, HP: Hexagonal Primitive  
 FCO: Face-centered orthorhombic. UD: Unidentified  
 Source: Author's own elaboration.

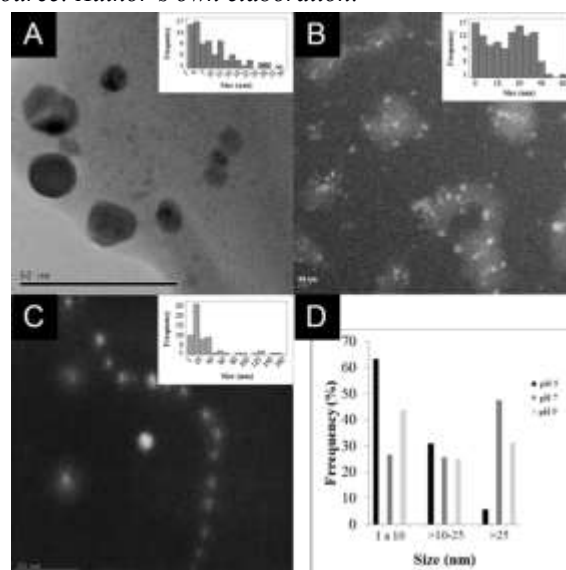


Fig.2: Low magnification electron micrographs of the Ag nanoparticles obtained at pH values: (A) 5; (B) 7; (C) 9; and the corresponding size distribution plots. The histograms show the range of particle size distribution (D).

The optimum pH was found to be 5, where over 60% of particles have sizes between 1 and 10 nm. These results are according to Baghizadeh et al. (2014), they report

optimum pH=4 to synthesized AgNPs with *P. harmala* seeds. However, Babu et al. (2014) using medicinal *Zizyphus xylopyrus* bark extract to obtain AgNPs they show surface plasmon resonance (SPR) peaks of AgNPs at neutral and alkaline conditions, which is in contrast to our results, this may be due to biomass type used.

Figure 3 shows the PSD of AuNPs, there was observed an influence by pH value, in Figures 3a and 3b the most nanoparticles were in 1-10nm with pH= 5 and 7, but in pH= 11 a majority of particles was in 20-60 nm. In Table 1 the size average was included for these nanoparticles,  $6.3 \pm 3.3$  nm,  $4.2 \pm 1.79$  nm and  $47.8 \pm 38$  nm for pH 5, 7 and 11 respectively. The main result was for neutral pH in which small-sized nanoparticles and the larger number of nanoparticles were formed; this is in according to the literature (Li, Li, Wan, Xu, & Hou, 2011).

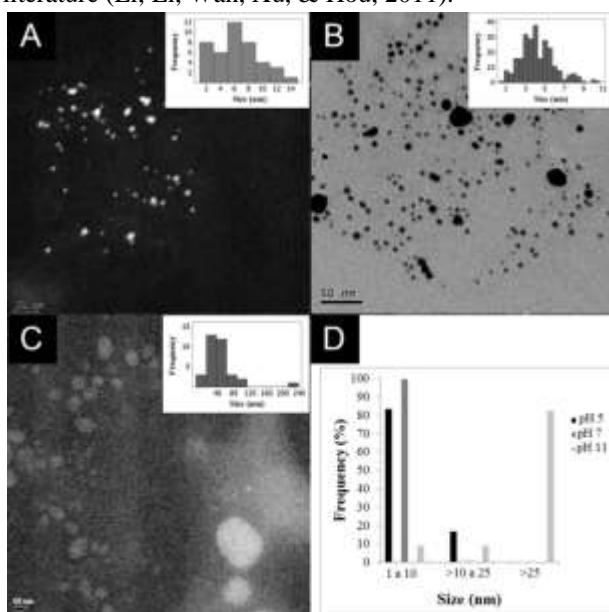


Fig.3: Low magnification electron micrographs of the Au nanoparticles obtained at pH values: (A) 5; (B) 7; (C) 11; the corresponding size distribution plots. The histograms show the range of particle size distribution (D).

HRTEM analysis. Significant results have been observed in PSD for AuNPs and AgNPs at different pH values, and the same characteristics were previously observed for other metals. (Rosano et al., 2006; Schabes et al., 2006; Canizal et al., 2006). Therefore, the optimal size distribution was obtained for AgNPs at pH=5 and for AuNPs at pH=7, where particles formed had small and regular size and symmetrical shape. Canizal et al. (2006) related the structure of the nanoparticles with the passivating agents, in this case, tannins from the biomass. To determine the crystal arrangement formed in the AgNPs and AuNPs were analyzed HRTEM images.

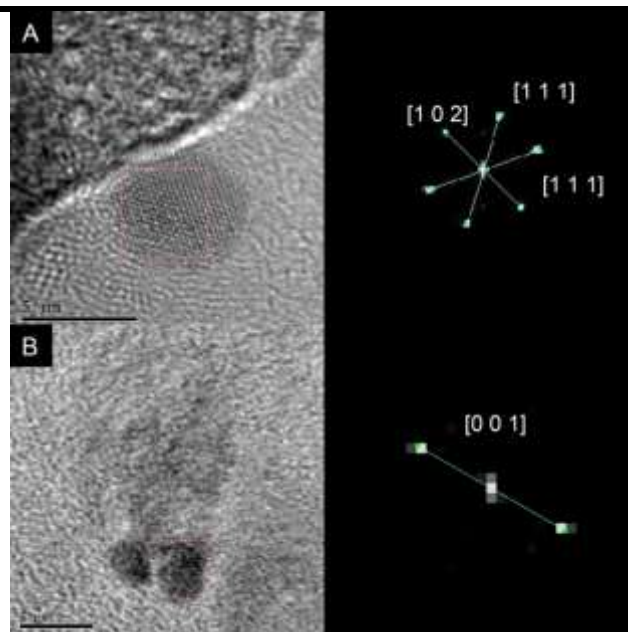


Fig.4: HRTEM images of particles observed in AgNP samples at (A) pH5 and (B) pH9, with their corresponding FFT for each image. (A) FCC nanoparticle at [111] and [102], (B)  $Ag_6O_2$  nanoparticle at the [001] orientation.

Figure 4 shows sets of comparable images for representative structures in AgNPs samples obtained at pH 5 and 9 with their corresponding FFT. For pH= 5 (Figure 4a) it was possible to identify that 86% of nanoparticles correspond to a face-centered cubic (FCC) structure with lattice fringes having a spacing of  $2.35 \text{ \AA}$  of interplanar space (JCPDS card 87-0720) and its growth preferentially over [111] and [102] planes. The rest of nanoparticles in the sample were not identified. In the case of pH = 7, the particles were formed by just a few atoms, so the contrast is not high. However, for pH = 9 (Figure 4b) it was observed that 80% of the nanoparticles corresponds to  $Ag_6O_2$  with a hexagonal structure (HP) and lattice fringes with a spacing of  $5.58 \text{ \AA}$  of interplanar space (JCPDS card 74-0878) in [001] orientation. According to with this results, it can be possible to establish, for pH=5 Ag nanoparticles were obtained, for pH=9 the silver oxide nanoparticles ( $Ag_6O_2$ ) was synthesized, the results were summarized in Table 1.

Figure 5 shows sets of comparable images for representative structures for AuNPs samples obtained at pH 5, 7 and 11 and their corresponding FFT. For pH= 5 it was possible to identify two AuNPs compositions. The first one (Figure 5a) 81% of nanoparticles with lattice fringes having a  $2.33 \text{ \AA}$  of interplanar space (JCPDS card 011172). It was according with the [111] d-spacing of bulk Au (Yin, Chen & Wu, 2010); the second one, 19% were of AuO (Figure 5 b) with lattice fringes having a  $2.22 \text{ \AA}$  of interplanar space (JCPDS card 23-0278). In the

case of the sample at pH=7 specimens, two structures were observed too.

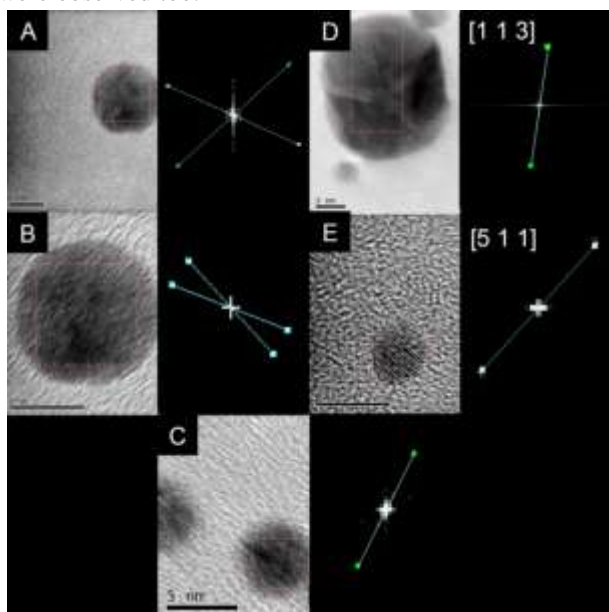


Fig.5: HRTEM images of particles observed in AuNP samples of (A & B) pH 5 and (C & D) pH 7, (E) pH 11 with their corresponding FFT for each image. (A) Au nanoparticle with [111] orientation, (B & C) AuO nanoparticles, (D) orthorhombic Au<sub>2</sub>O<sub>3</sub> nanoparticle with [133] orientation, (E) orthorhombic Au<sub>2</sub>O<sub>3</sub> nanoparticle with [511] orientation.

In the first one 68% of the sample corresponds to AuO nanoparticles (Figure 5c); with lattice fringes having a 2.22 Å of interplanar space (JCPDS card 23-0278). Moreover, in the second one, 32% of nanoparticles were in Au<sub>2</sub>O<sub>3</sub> composition with a spacing of 1.19 Å (Figure 5d) corresponding to a face-centered orthorhombic (FCO) structure (JCPDS card 43-1039) in orientation [133]. Finally, for pH=11 also an Au<sub>2</sub>O<sub>3</sub> nanoparticle in FCO structure (90%) was observed (Figure 5e) with lattice fringes having a spacing of 2.15 Å of interplanar space (JCPDS card 43-1039) on the [511] plane. According to these results, to obtain Au<sup>0</sup> or Ag<sup>0</sup> the best value for pH was 5, 7 respectively, while for AgNPs only one oxide was identified for particles (Table1), for AuNPs two oxides were recognized, only one configuration of gold oxide is clearly associated with pH= 11 (Au<sub>2</sub>O<sub>3</sub>). The results are condensed in Table 1.

The composition of the nanoparticle surface is intimately related to their final application because the surface will be the first aspect experiences either the environment or the organism (Christian, Von der Kammer, Baalousha & Hofmann, 2008). The metallic nanoparticles produced by biosynthesis were silver or gold oxide in the most cases, and they could be used in industrial and biomedical applications due to antimicrobial, photocatalytic, and luminescent properties.

Changes in the size distribution and structure of AgNPs and AuNPs are due to the pH varying. The AgNPs at pH= 5 was obtained the smallest size; the reduction is slow and efficient, these nanoparticles were well passivated because they search of a minimum energy surface. However, for AuNPs the smallest size was reached at pH 7, so is possibly a fast reduction process that reduces the ions faster causing aggregation and different configurations. Also, might be due to a weak passivation of tannins in the biomass with Au (Canizal, et al., 2006).

## 5. CONCLUSIONS

This study proposed an eco-friendly, nonhazardous, cheap, and rapid method for AgNPs and AuNPs synthesis using an invasive aquatic plant, water hyacinth, which it converts a pest to a reduction agent and a value-added chemical. The TEM analysis revealed the particle size distribution for each pH (acid, neutral or alkaline) which seems to be trimodal. For AgNPs, at pH= 5 was determined the highest percentage of nanoparticles with sizes in the range (1 a 10 nm) with a structure cubic face-centered. In the case of AgNPs, it was observed that the size and structure of nanoparticles were strongly dependent on the pH value. For AuNPs, at pH= 7 the smallest and less dispersed were obtained with two configurations (AuO and Au<sub>2</sub>O<sub>3</sub>). Metallic nanoparticles obtained could be applied in industry and biomedicine. Bioreduction method lets it obtained silver and gold nanoparticles, and the use of an invasive plant makes it a sustainable method to synthesise metallic nanoparticles.

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