

Full Length Research Paper

Unexplored vegetal green synthesis of silver nanoparticles: A preliminary study with *Corchorus olitorus* Linn and *Ipomea batatas* (L.) Lam

François Eya'ane Meva^{1,2*}, Marcelle Loretta Segnou¹, Cecile Okalla Ebongue^{3,4}, Agnes Antoinette Ntomba⁵, Djiopang Yadou Steve¹, Fanny Aimee Essombe Malolo¹, Lidwine Ngah¹, Harouna Massai⁶ and Emmanuel Mpondo Mpondo^{1,7}

¹Department of Pharmaceutical Sciences, Faculty of Medicine and Pharmaceutical Sciences, University of Douala, P.O. Box 2701, Douala, Cameroon.

²Department of Chemistry, Faculty of Sciences and Engineering, University of Hull, HU6 7RX, Hull, United Kingdom.

³Department of Biological Sciences, Faculty of Medicine and Pharmaceutical Sciences, University of Douala, P.O. Box 2701, Douala, Cameroon.

⁴Clinical Biology Laboratory, General Hospital of Douala, P.O. Box 4856, Douala, Cameroon.

⁵Department of Animal Biology and Physiology, Faculty of Science, P.O. Box 24157, University of Douala, Cameroon.

⁶Department of Chemistry, University of Maroua, P.O. Box 55, Maroua, Cameroon.

⁷Department of Pharmacotoxicology and Pharmacokinetics, University of Yaounde I, P.O. Box 1364, Yaounde, Cameroon.

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Antibacterial properties of silver ion are known from ancient times. The plant extract mediated synthesis of nanoparticles is gaining popularity due to green chemistry for the generation of nanosized materials. *Corchorus olitorus* Linn and *Ipomea batatas* (L.) Lam are world crops having leaves of high nutritional value. In the present work, Ag and AgCl nanoparticles were synthesized by incubating leaf aqueous extracts with silver nitrate salt, making them to react in the dark. Plasmon resonance bands, studied by ultraviolet spectroscopy, have been obtained. X-ray diffraction (XRD) revealed the average size of pure crystallites composed of Ag and AgCl. Optimization studies for the synthesis process highlight positive impact of pH, reaction time and reactants quantities.

Key words: Silver, nanoparticles, *Corchorus olitorus*, *Ipomea batatas*, X-ray diffraction.

INTRODUCTION

Silver nanoparticles are receiving great attention due to their applications in different areas such as biotechnology,

packaging, electronics, medicine and coatings (Solgi, 2014). The synthesis of nanoparticles using bio-entities

*Corresponding author. E-mail: mevae@daad-alumni.de.

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presents more advantages over other conventional processes due to the availability of more biological components for their formation. The rich diversity of such entities should be explored for the synthesis of bionanomaterials (Nahar et al., 2015).

Traditionally, silver metal is used to control pathogenic infections and prevent food spoilage. Silver is used as wound healing agent and for ulcer treatment (Singh et al., 2013). In fact, now-a-days, the colloidal silver nanoparticles have been used as antimicrobial agent, wound dressing material, bone and tooth cement and water purifier as well (Narayanan and Sakthivel, 2011). Most of the nanoparticles used in biotechnologies rely on the use of toxic reducing agents (e.g. sodium borohydride) and harmful organic solvents (e.g. N, Ndimethylformamide (DMF), tetrahydrofuran (THF)) (Nahar et al., 2015). These chemicals have potential biological and environmental risks. To solve these problems, biosynthesis of nanomaterials is a growing need to develop environmental-friendly and clean green methods of nanoparticles preparation using non-toxic chemicals and renewable reducing agents (Nahar et al., 2015).

Plant crude extract contains novel secondary metabolites such as phenolic acid, flavonoids, alkaloids and terpenoids. These compounds are mainly responsible for the reduction of ionic into bulk metallic nanoparticles formation (Aromal and Philip, 2012). These primary and secondary metabolites are constantly involved in the redox reaction to synthesize eco-friendly nanosized particles (Kuppusamy et al., 2014).

Corchorus olitorus Linn (Jute), a vegetal from family Tiliaceae, is a native plant of tropical Africa and Asia, and has since spread to Australia, South America and some parts of Europe. It's a leafy vegetable popularly used in soup preparation and as a food source in Asia, the Middle East, and parts of Africa. In addition to adding a distinct flavor to food, jute leaves also have nutritional value, and act as thickeners in soups, stews and sauces (Islam, 2013). Together with other leafy species, *Corchorus olitorus* is the main source of dietary protein in many tropical countries (Tulio et al., 2002).

The plant has uses in folk medicine for the treatment of fever, chronic cystitis, cold and tumours. The plant extracts exhibit antioxidant capacity, possess reducing power and OH scavenging ability. The additive/synergistic antioxidant activities of the hydrophilic and lipophilic constituents may contribute to the medicinal properties of *C. olitorius* L. leaf (Obboh et al., 2009). The phytochemical screening of the leaves of *C. olitorus* showed the presence of alkaloids, cardiac glycoside, flavonoids, phenols, saponins, tanins, terpenoids and steroids (Hamzah et al., 2014; Olajire and Azeez, 2011). Jute also contains high level of all essential amino acids except methionine (Tulio et al., 2002).

Ipomoea batatas (L.) Lam. (sweet potato) is a vegetable, in the family Convolvulaceae and highly

nutritious (Srisuwan et al., 2006; Mbaeyi-Nwaoha and Emejulu, 2013). The species is an important food crop widely grown in tropical, subtropical and warm temperate regions (Srisuwan et al., 2006; Mbaeyi-Nwaoha and Emejulu, 2013).

Sweet potato is very rich in a lot of nutrients which include: carbohydrate, vitamins and minerals, also it contains much higher levels of provitamins A, vitamin C and minerals than rice or wheat (Villanueva, 1977). Leaves have been evaluated to show appreciable amounts of zinc, potassium, sodium, manganese, calcium, magnesium, iron, vitamin A, B2, B6, C and E, and fiber (Antia et al., 2006). The phytochemical screening showed positive results for triterpenes/steroids, alkaloids, anthraquinones, coumarins, flavonoids, saponins, tannins and phenolic acids. The plant has an antioxidant capacity (Padda and Picha, 2008). Six compounds were isolated from 90% ethanol extract and identified as tetracosane, myristic acid, beta-sitosterol, beta-carotene, daucosterol and quercetin (Lv et al., 2009). The leaves contain low levels of cyanide, phytic acid and tannins. They are rich in unique phytonutrients, including polysaccharide related molecules called batatins and batatosides (Low et al., 2009).

Lyophilized leaf powder from sweet potato strongly suppressed the growth of food poisoning bacterial such as *Staphylococcus aureus*, *Bacillus cereus*, *Escherichia coli* O157 (Islam, 2006; Yoshimoto et al., 2001). The leaf decoction is used as folk medicine to remediate tumors in the mouth and throat. This is a folk remedy for asthma, bug bites, burns, catarrh, ciguatera, convalescence, diarrhea, dyslactea, fever, nausea, renosis, splenosis, stomach distress, whitlows, burning sensations, constipation, general weakness, renal calculi, sexual stimulant, jaundice and ulcer (Osime et al., 2008). The leaves are also used in the treatment of hookworm, hemorrhagic and abscesses, while the tuber is used for the treatment of asthma (Udoh et al., 2010). The leaves also possess anti-hyperglycemic activity (Li et al., 2009) and used in the treatment of diabetes (Islam, 2006). It is also a suitable substrate for the efficient production of biofuel ethanol using *Saccharomyces cerevisiae* (Saritha et al., 2010) and has potential for other uses such as fuel and animal feed in which the leaves and vines are used for feeding rabbits, sheep, goats and cattle (Okungbowa and Osagie, 2009). Using *Ipomoea aquatica* (Convolvulaceae) leaf extract, silver nanoparticles were reported to be synthesized (Sivaraman et al., 2013).

In view of their importance and based on their chemical composition, *C. olitorus* and *I. batatas* can be used for the green synthesis of silver nanoparticles. The main aim of this research work was to highlight the green bioreduction of ionic silver to nanoparticles using ultraviolet-visible spectroscopy. A study of the crystallinity, phase and composition of the nanoparticles using X-ray diffraction is presented. Furthermore, synthetic optimizations varying pH, incubation conditions, extract



Photograph 1. *Corchorus olitorus* (left) and *Ipomea batatas* (right).

and silver ion quantities are discussed.

MATERIALS AND METHODS

Silver nitrate (AgNO_3) was obtained from Sigma-Aldrich chemicals Germany, H_2SO_4 (98% from Merck KGaA, Darmstadt, Germany) and NaOH (R. P. Normapur Prolabo Paris) and used as received. De-ionized water was used throughout the reactions. Fresh leaves of *Corchorus olitorus* Linn (Tiliaceae) and *Ipomea batatas* (L.) Lam (Convolvulaceae), were procured from local market, Douala, Cameroon, and identified at the national herbarium of Cameroon by TADJOUTEU Fulberg under number of deposit 14725/SRF Cam and 26429/SRF Cam respectively. All glass wares were washed with dilute nitric acid (HNO_3) and de-ionized water, and then dried in hot air oven.

Instrumentation

The formation of silver nanoparticles (Ag-NPs) was observed by measuring the UV-Vis spectrum of 2.5 mL of the reaction suspension at different time intervals. If absorbance was higher than 4.5 u.a., the sample was diluted by a factor of $\frac{1}{2}$ with distilled water. An UV-visible Uviline 9100 spectrophotometer (single beam, halogen light source) operated at 1 nm resolution, optical length of 10 mm with a measuring range from 320 to 1100 nm was used. UV-visible analysis of the reaction mixture was observed for a period of 300s. XRD measurements were carried out using a PANalytical Empyrean Serie 2 X-ray diffractometer (Cu K-Alpha1 [\AA] 1.54060, K-Alpha2 [\AA] 1.54443, K-Beta [\AA] 1.39225) by preparing a thin film on silicium substrate.

Preparation of aqueous extract

Aqueous extract of *C. olitorus* and *I. batatas* were prepared using 10 g of freshly collected leaves (Photograph 1). The leaves were surface cleaned with running tap water, followed by distilled water and boiled with 200 mL of distilled water at 80°C for 5 min. The extract was filtered through Whatman paper No. 1 and stored at 4°C for further use, being usable for 1 week due to the gradual loss of plant extract viability for prolonged storage.

Green synthesis of silver nanoparticles

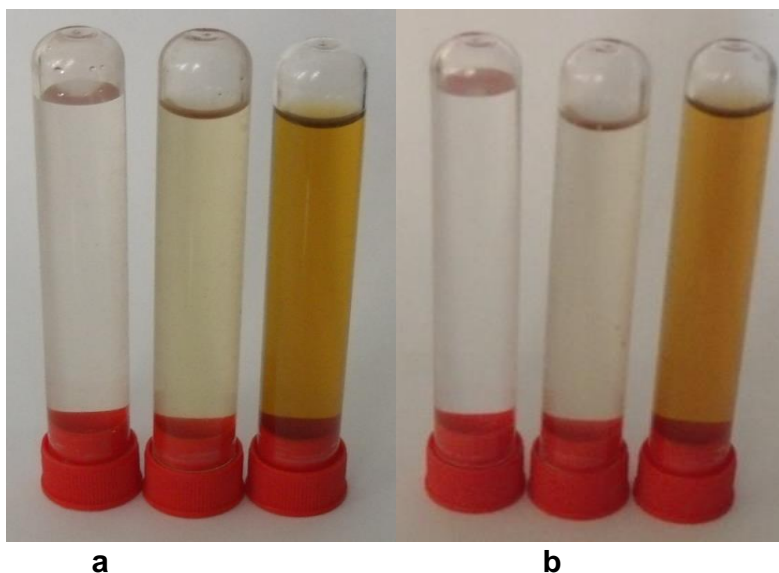
For the synthesis of the silver nanoparticles, a volume of plant leaf extract (5, 10 and 15 mL) was added to 50 ml of 10^{-3} , 10^{-2} or 10^{-1} M aqueous AgNO_3 solution and incubated at room temperature in the dark to minimize the photoactivation of silver nitrate under static conditions. To study the synthesis of nanoparticles, 2.5 mL aliquots were taken and absorbance was measured after 5, 10, 20, 30, 40, 50 and 60 min of the beginning of the reaction. Different pH gradients were used for optimal time to obtain silver nanoparticles. The pH of the solutions was adjusted using 0.1 N H_2SO_4 and 0.1 N NaOH solutions. The contact time of incubation for all studied samples was varied from 1 to 24 h and then 96 h.

RESULTS AND DISCUSSION

Ultraviolet-visible Ag nanoparticles formed and incubation contact time

The absorption spectra of the synthesized silver nanoparticles were recorded against water in order to monitor the formation and stability of silver nanoparticles. Visual observation was made and first hour of reaction was recorded. Silver nitrate solution, plant leaf extract and silver nanoparticles solutions are shown in Photograph 2 [*C. olitorus* (a) and *I. batatas* (b)]. The evolution of the surface plasmon absorbance bands during the synthesis of silver nanoparticles using 50 mL of AgNO_3 10^{-3} M with 10 mL of extract concentration during the first hour is shown in Figures 1 and 2. The solution color change to pale yellow within seconds, and then to yellowish brown, indicates the formation of silver nanoparticles.

The evolution of the surface plasmon absorbance bands during the synthesis of silver nanoparticles using 5, 10 and 15 mL of leaf extract, and 50 mL of different concentrations of AgNO_3 (10^{-3} M, 10^{-2} M, 10^{-1} M) is shown in the chart supplement 1 (*C. olitorus*) and



Photograph 2. Silver nitrate solution (left), plant leaf extract (middle) and silver nanoparticles solutions (right). a. *Corchorus oleratus* leaf extract; b. *Ipomea batatas* leaf extract.

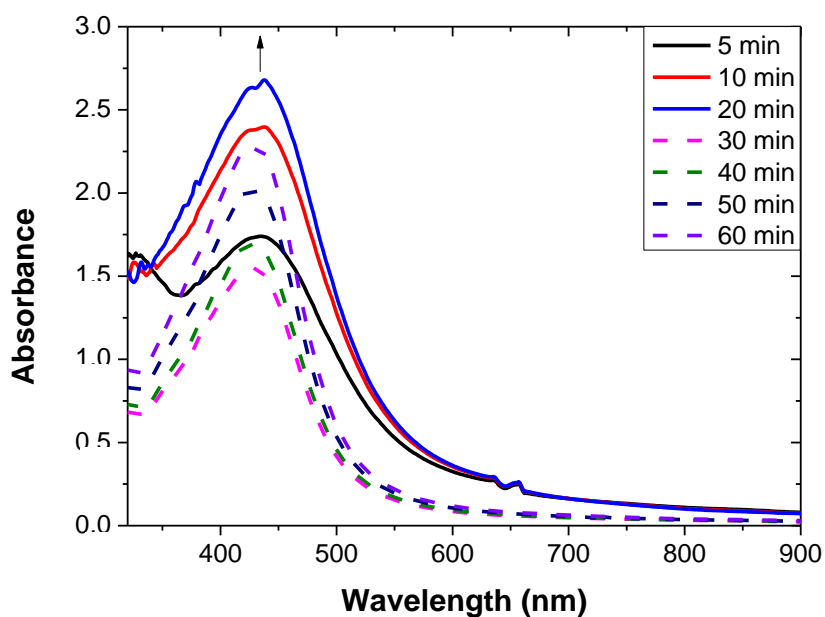


Figure 1. Monitoring 60 min of the reaction between 10 mL of *Corchorus oleratus* leaves extract and 50 mL AgNO_3 10^{-3} M. Dash curves represent solutions diluted by 0.5 due to their maxima higher than the maximum 4.5 u.a.”

supplement 2 (*I. batatas*). The visible spectra had been observed after 1, 24 and 96 h of incubation time in the dark to eliminate silver ions photoactivation. It is notable that the production of silver nanoparticles increases with incubation time which tends to produce polydispersed silver nanoparticles. More leaf extract tend to produce

polydispersed and aggregated solution, a situation already found using *I. aquatica* leaf extract in 1:1 volume ratio of extract: silver ion (Sivaraman et al., 2013). At 10^{-1} M concentration of the silver ions with the different *C. oleratus* and *Ipomea batatas* extract concentrations, the nanoparticles exhibited aggregation because of the

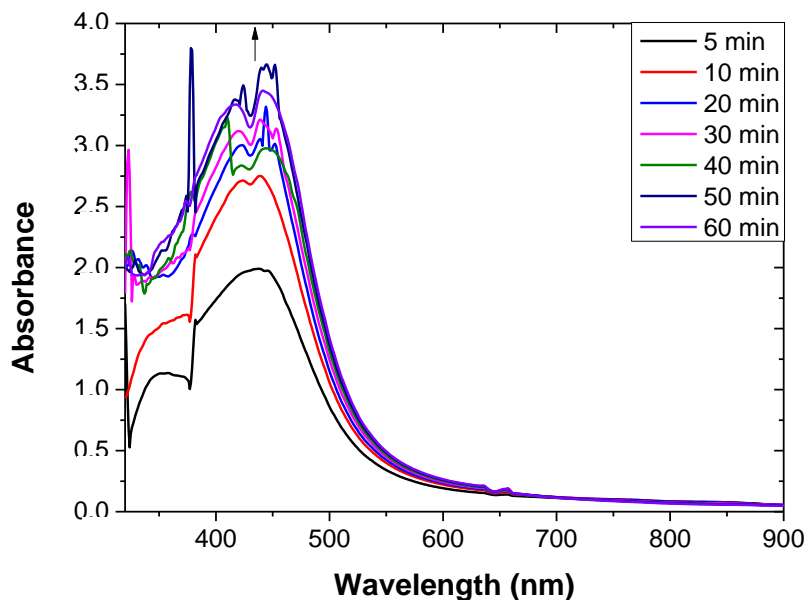


Figure 2. 60 min monitoring of the reaction between 10 mL of *Ipomea batatas* leaves extract and 50 mL AgNO_3 10^{-3} M.

deficiency of molecule of leaf extract to act as protecting agents. The barrier potential developed as a result of the competition between weak van der Waals forces of attraction and electrostatic repulsion is broken (Prathna et al., 2011). It is notable that during the first hour of incubation, silver nanoparticles mediated *C. olitorus* are monodispersed AS compared to the situation of silver nanoparticles mediated *I. batatas* leaf extract. The biomolecules present in the two plant leaves extracts act as reductant as well as capping agent, therefore mediate the synthesis as well as stabilization of the silver nanoparticles. The surface plasmon resonance band increases with silver ion concentration as observed for olive leaf extract (Khalil et al., 2013). It can be seen that the absorbance band maxima of Ag-nanoparticles using *C. olitorus* and *I. batatas* is in the range 400-450 nm due to surface Plasmon resonance (Mulvaney, 1996) of Ag-nanoparticles. The UV-Vis spectra also revealed that the formation of Ag-nanoparticles occurred rapidly within few minutes, indicating that both extracts rapidly increase the biosynthesis of silver nanoparticles.

Effect of pH

UV-visible spectra recorded during the formation of silver nanoparticles from pH 2 to 12 are shown in Figures 3 (*C. olitorus*) and 4 (*I. batatas*). The nanoparticle solution color at each pH studied is shown in Photographs 3 (*C. olitorus*) and 4 (*I. batatas*). It can be seen that plasmon absorbance bands increase with increasing pH from 2 to 12, which can be due to the increase in production of colloidal silver nanoparticles and reduction rate. The absorbance does not decrease at pH higher than 8 as it

was obtained by other investigators for olive leaf extracts (Khalil et al., 2013). Acidic pH of 6 is enough to reduce ionic silver for both plant extracts. In the case of *I. batatas* (L.) Lam mediated synthesis, at pH 6, the nanoparticles are already polydispersed and slightly aggregate. Furthermore, it is observed that the brown color of the nanoparticles appeared shortly after mixing the AgNO_3 with both extracts at pH 6 to 12. Irvani and Zolfghari (2013) observed that pH affects the amount of nanoparticle production and their stability when they studied nanoparticle production and their stability, using *Pinus eldarica* bark extract.

X-ray diffraction

The typical powder XRD patterns of the prepared nanoparticles are shown in Figures 5 and 6. These patterns are compatible with the cubic phase of Ag with diffraction points at 2θ values of 38, 44.2, 64.6 and 76.7° and can be indexed to the (111), (200), (220) and (311) planes of the face centered cubic (FCC) structure, respectively (JCPDS file: 65-2871). The XRD pattern also showed the presence of the cubic phase of AgCl at 2θ values of 27.8°, 32.2°, 46.2°, 54.8° and 57.4°, corresponding to the (111), (200), (220), (311) and (222) planes, respectively (JCPDS file: 31-1238). The average crystallite size of the synthesized NPs was determined using the Debye-Scherrer equation:

$$Dv = K\lambda/\beta \cos \theta$$

where Dv is the average crystalline size; K is a dimensionless shape factor, with a value close to unity

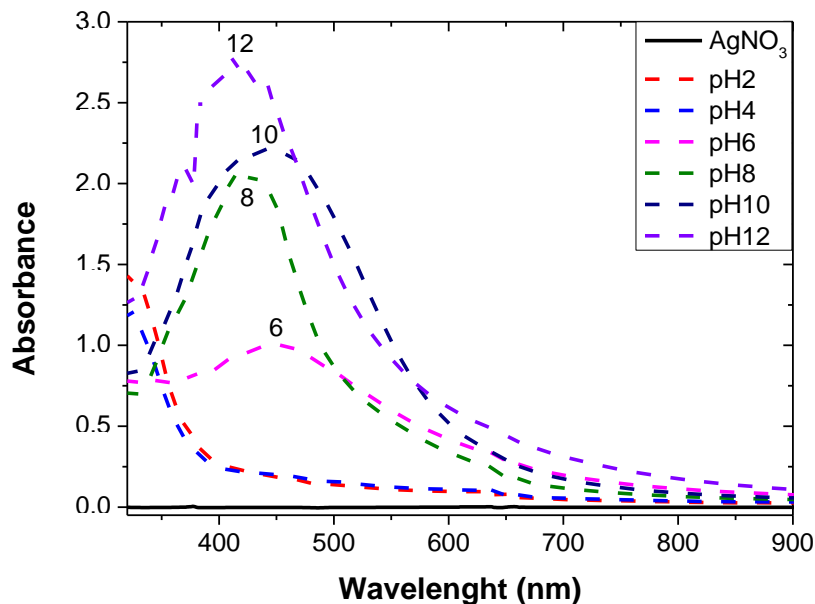


Figure 3. UV-Vis spectra of the variation of pH in *Corchorus olitorus* leaf extract. Dash curves represent solutions diluted by 0.5.

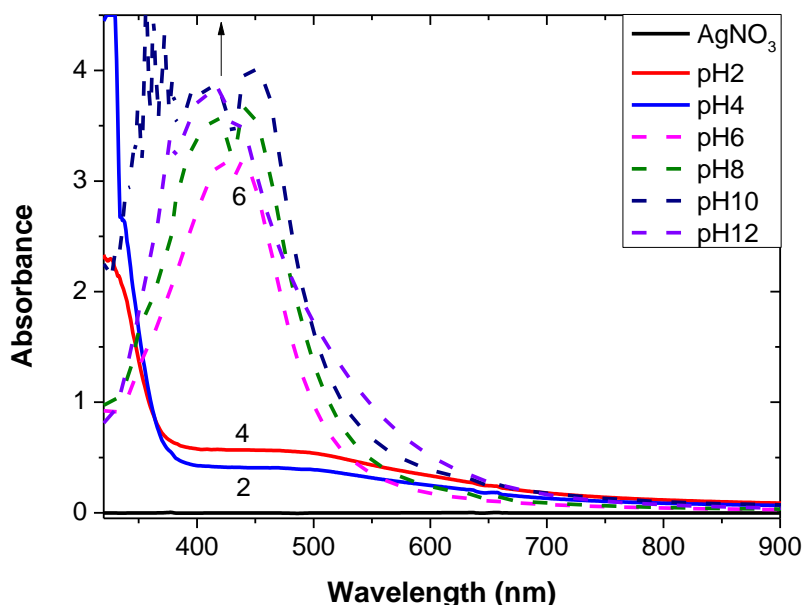
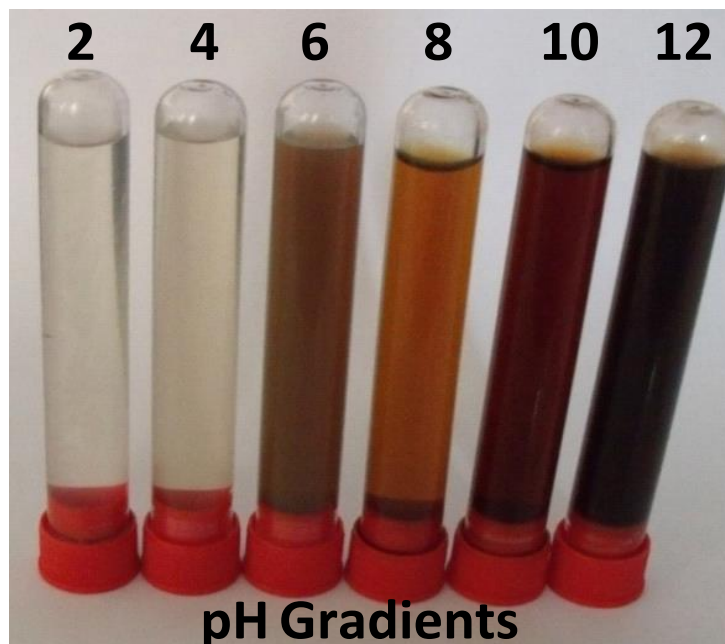


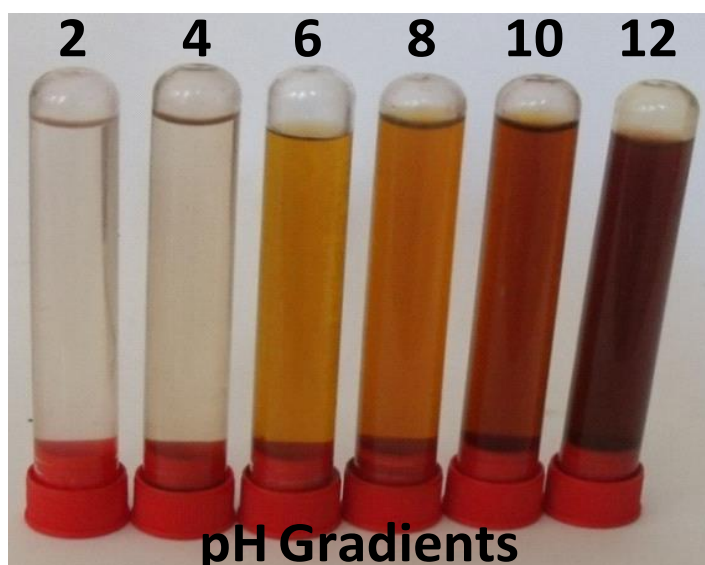
Figure 4. UV-Vis spectra of the variation of pH in *Ipomea batatas* leaf extract. Dash curves represent solutions diluted by 0.5.

(0.9); λ is the wavelength of Cu K α ; β is the full width at half-maximum of the diffraction peaks, and θ is Bragg's angle. No other characteristic peaks were found in the XRD spectra, indicating the high purity of the as-prepared Ag and AgCl nanoparticles. To calculate the average crystalline particle size of the synthesized Ag and AgCl nanoparticles, the most intense peaks of Ag and AgCl

were preferred. The (111) and (200) lattice planes of Ag and AgCl were have selected to calculate the average crystalline particle size of Ag and AgCl NPs. The calculated average crystalline particle size of the Ag and AgCl *Ipomea* was found to be 67.3 nm and 37.9 nm for Ag and AgCl, respectively. Whereas 30 and 37.9 nm where found for Ag and AgCl crystallites of the Ag and



Photograph 3. Colors of Ag nanoparticles solution at pH 2, 4, 6, 8, 10 and 12 for *Corchorus olitorus*.



Photograph 4. Colors of Ag nanoparticles solution at pH 2, 4, 6, 8, 10, and 12 for *Ipomea batatas*.

AgCl *Corchorus* NPs.

Pattern identification and quantification shows the formation of pure crystals with Ag55% AgCl45% composition for Ag-*Ipomea* and Ag44%AgCl56% for Ag-*Corchorus*. The intense and narrow diffraction peaks revealed the crystalline nature of the synthesized nanoparticles (Wang et al., 2010).

Conclusions

The use of plants for green synthesis of nanoparticles is gaining more popularity over the years. The synthesis is cost-effective, eco-friendly, efficient and rapid. *C. olitorus* Linn and *I. batatas* (L.) Lam are important crops for the world consumption. Both leave extracts are capable of

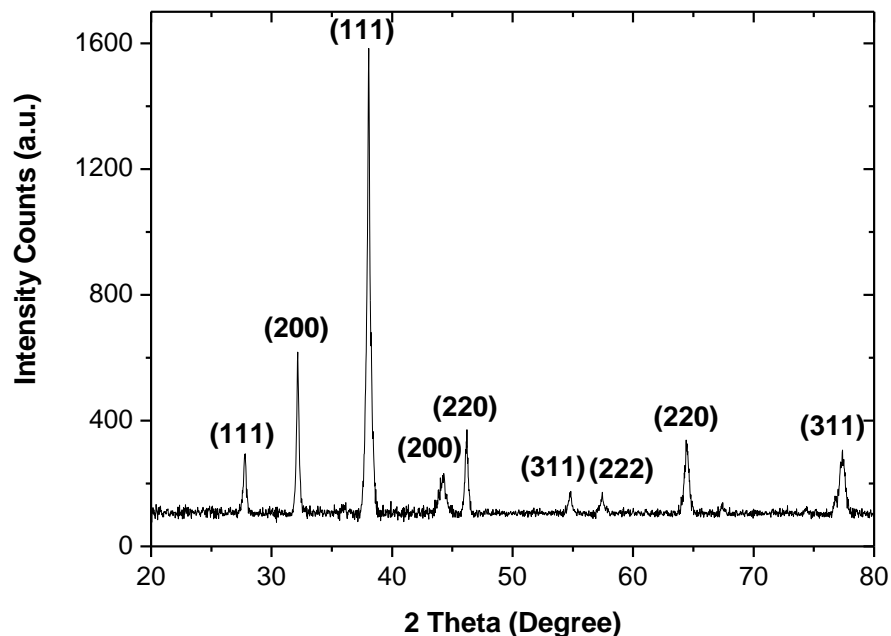


Figure 5. XRD pattern of the nanoparticles in *Ipomea batatas*.

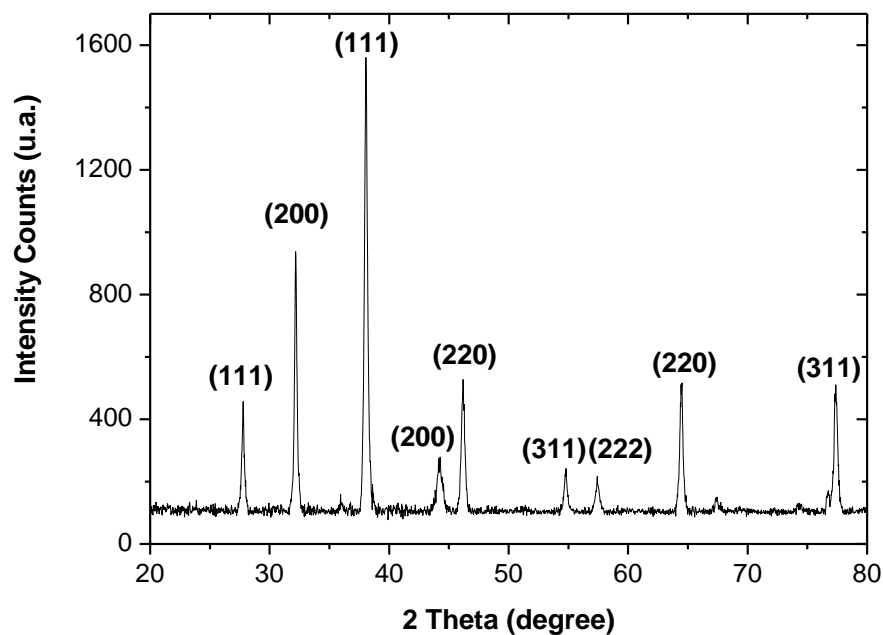


Figure 6. XRD pattern of the nanoparticles in *Corchorus oleratus*.

reducing ionic silver to nanosilver within 5 min. Synthetic optimizations with different incubation conditions, reactants and pH could be followed by ultraviolet-visible spectroscopy. The measurements show that the synthesis is promoted even at acidic pH of 6. The plasmon resonance bands intensity (between 400 and 450 nm) increases with extracts and silver ion quantity.

Powder X-ray diffraction studies confirm that the pure nature of the crystallites is composed of Ag and AgCl nanoentities with size of 67.3 and 37.9 nm for Ag and AgCl, respectively for Ag/AgCl-*Ipomea* and 30 and 37.9 nm for Ag/AgCl-*Corchorus*. The synthesized silver nanoparticles can find applications in different areas such as medicinal and water treatment.

Conflict of interests

The authors have not declared any conflict of interests.

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