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EXPLORING PH EFFECTS ON STABILITY, SIZE, AND SHAPE OF GOLD NANOPARTICLES SYNTHESIZED USING AKASHMONI POD EXTRACT

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ABSTRACT
An eco-friendly and rapid method has been developed to prepare gold nanoparticles (Au NPs) using an aqueous extract of Akashmoni (*Acacia auriculiformis*) pods without adding any external reducing agent. The formation of Au NPs is confirmed by the colour change and UV–Visible Absorption spectroscopy. TEM, XRD, and zeta-potential studies confirm their nanoscale size, crystalline nature, and stability. The study shows that pH has an important effect on nanoparticle formation, shape, and stability, with better stability at pH ≥ 9.5. The results show that Akashmoni pod extract can act as a natural reducing and stabilizing agent, making the synthesis simpler and more environmentally friendly. This work can support future research on the large-scale preparation and biological and catalytic applications of plant-based nanoparticles. The study contributes to SDG-12: Responsible Consumption and Production by reducing the use of conventional chemical reagents. Therefore, this method provides a simple green chemistry approach for sustainable nanomaterial production utilising locally available plant resources.
Keywords: Gold Nanoparticles; Eco-Friendly; Biosynthesis; Akashmoni
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INTRODUCTION

Researchers now study metal nanoparticles far more than before because these tiny particles show different optical, chemical, photoelectrochemical, and electronic behaviour than their bulk metal analogues.[1,2] Synthesis of metal nanoparticles using traditional wet chemical methods includes the use of ionizing radiation, hazardous chemicals such as sodium borohydride, as well as non-aqueous solvents such as ethylene glycol.[3] Recently, various scientists have been synthesizing controlled-size and shape metal nanoparticles by using a "green" approach at ambient temperature and pressure. The method of synthesizing metal nanoparticles using plant extracts is cheap and therefore can be used as a cost-effective and suitable substitute for the scale-up synthesis of metallic nanoparticles. In recent years, metal nanoparticles have typically been obtained by reducing the metal ions with plant biomolecules such as flavonoids, amino acids, enzymes, and polysaccharides.[4] The synthesis of Ag, Cu, and Au nanoparticles using broth extracts of numerous plants is also found to be efficient and rapid.[5–9] Akashmoni's botanical name, *Acacia auriculiformis,* is a fast-growing tree commonly well known for its medicinal properties.[10] Research shows it holds tannins, terpenoids, and a range of polyphenols.[11] In previous work, we reported the silver nanoparticle preparation using aqueous extract of pods of the plant Akashmoni.[12] The present study supports sustainable nanomaterial production by using a simple, rapid, and eco-friendly method to prepare gold nanoparticles from Akashmoni water extract without adding any external reducing agent. This approach reduces the use of chemicals and promotes greener and more responsible use of resources as per SDG 12 – Responsible Consumption and Production.

EXPERIMENTAL

Collection of Plant Material

Pods of Akashmoni plant were obtained from Konkan region of Maharashtra, India between the latitude 220 1" to 160 4" N and longitude 720 6" to 8009" E as and when required. Tetrachloroauric acid and Sodium hydroxide (Sigma), methanol (Merck) were used without further purification. All syntheses were

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carried out using 18.2 MΩ electrical resistivity purified water. The glassware was cleaned with chromic acid and dried in an oven at 107°C.

Preparation of Plant Extract

Plant extract of *pods* was prepared by boiling about 5 gm of the pods in 50 mL of Millipore water for 7 minutes. After cooling at room temperature, it was filtered through Whatman filter paper No. 1 and stored at 4°C.

Biosynthesis of Gold Nanoparticles

Synthesis of gold nanoparticles was carried out by mixing 2 mL of aqueous extract with 8 mL aqueous solution of $HAuCl_4$ at room temperature at the inherent pH, the final concentration of the gold in the solution is $5x10^{-4}$ M. To study the effect of pH on the formation of gold nanoparticles $1x10^{-3}$M NaOH was used as per the pH requirement. The observed changes in color of nanoparticles were monitored using UV-visible spectroscopy.

Characterization

Chemito Spectra-scan UV 2600 instrument was used for recording UV- Vis absorption spectra. Zetasizer from Malvern was used to record zeta potential. Phillips Analytical (PW 1729) diffractometer employing Cu Kα radiation (1.5406 Å) model was used to study X-ray diffraction patterns of the powder sample. Phillips CM 200 electron microscope instrument was used for TEM characterization. TEM micrographs obtained were used to calculate the average size of nanoparticles.

RESULTS AND DISCUSSION

Characterisation of Gold Nanoparticles

After mixing tetrachloroauric acid with Akashmoni extracts while maintaining the inherent pH at 27°C, the color of the extract changed from yellow to purple, thus confirming the formation of gold nanoparticles.[3] The experiment was optimized by varying the concentration of gold ion precursor from 1x10-4 M to $1x10^{-3}$ M; the representative case for studying the kinetics of gold nanoparticle formation is depicted in Fig.-1. The reduction reaction seems to be fast and shows roughly linear growth in gold ion reduction, which gets saturated after 15 min (Fig.-1).

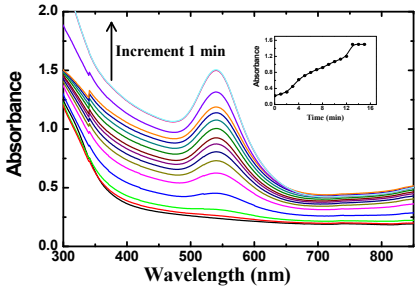


Fig.-1: UV-visible Absorption Graph of Gold Nanoparticles after Addition of Extract to 5 x 10^{-4} M $HAuCl_4$ at pH 2.2 (in 1cm path length cell). Inset: Evolution of Absorbance Yield Due to Gold Nanoparticles

pH Effect on Stability, Development and Shape of Gold Nanoparticles

Figure-2 depicts the pH effect on the development of gold nanoparticles. It is noticed that with increasing pH, the surface plasmon resonance band of the gold nanoparticles shifts towards the blue region. However, at pH 5.9, it shows a red shift, which again follows a blue shift as the pH was increased (Fig.-2). The nanoparticles' colour was also found to change accordingly, from pinkish magenta to wine red as pH increased from 2.2 to 10.5. Except at pH 5.9, it was brinjal violet. The adsorption of extract over Au nanoparticles might be the reason behind the observed shift in the spectrum.[3]

Figure-3 shows the photograph of gold sol prepared at different pHs. It was also observed that under basic condition ≥ pH 9.5, the synthesized gold nanoparticle sol showed enhanced colloidal stability. The stability of gold sols obtained at pH 9.5 and 10.5 is greater than one month without any precipitation which is further confirmed by zeta (ζ) potential measurement. The technique mostly used to check the stability colloids.[13,14]

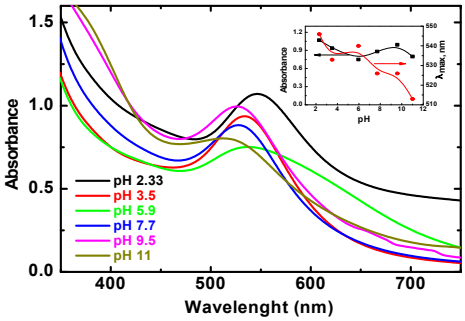


Fig.-2: UV-visible Spectrum of Gold Nanoparticles at Various pH range. Inset: Plot of O.D. and λ_{max} Vs pH of Au Nanoparticles

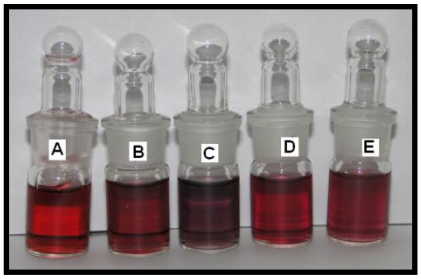


Fig.-3: Photograph of Gold Nanoparticles Obtained at different pH (A) 10.5, (B) 7.7 (C) 5.9 (D) 3.3 and (E) pH 2.2 (Inherent)

The ζ potential value is ≥ 30 mV considers as stable colloid.[14] The ζ potential values of gold sols prepared at different pHs is depicted in Fig.-6. Representative case of the zeta potential (ζ) plot for pH 10.5 is depicted in Fig.-4. The Zeta potential decreases at low pH, which leads to decrease in electrostatic repulsion between the particles.

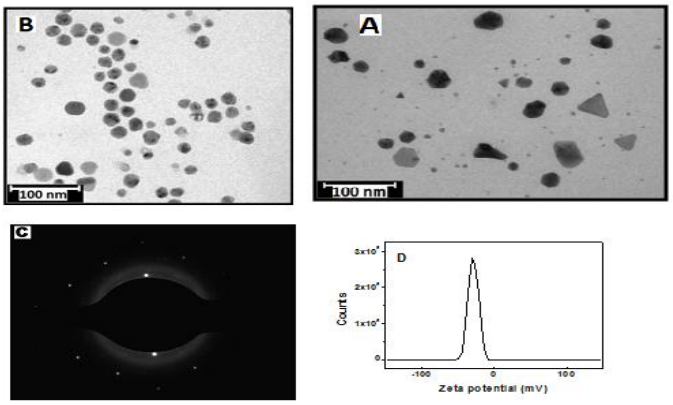


Fig.-4: Transmission Electron Microscopy Graph of Gold Nanoparticles (A) pH 2.2 (Inherent), (B) pH 10.5 and (C) Electron Diffraction Pattern at pH 2.2. (D) Zeta Potential of Gold Nanoparticles Prepared at pH 10.5

XRD in Fig.-5 shows the XRD pattern of the as-prepared gold nanoparticles, the patterns exhibit peaks at 38.2, 44.5, and 64.7 reveals the diffraction from (111), (200), and (220) planes of metallic gold (Ref. Code. 04-0784). The XRD peaks show broadening which is due to nanocrystalline nature of the sample. The particle size was calculated using Scherrer formula and was found to be ~23 nm.

The present work demonstrates an eco-friendly method for gold nanoparticle formation with extract of the *Akashmoni* plant. The method is optimised by varying the precursor conc., time, and pH. The reduction of gold nanoparticles starts after adding hydrochloric acid to the pods extract, as evidenced by a light yellow-to-wine-red color change.[3] After an optimum time for the formation of stable gold nanoparticles, there is no change in the absorption spectra was observed, showing the completion of reaction. Gold nanoparticle formation also depends on the optimum amount of metal precursors and the reducing agents

present in extract. The yield of nanoparticles therefore depends on the availability of metal ions in the reaction medium.

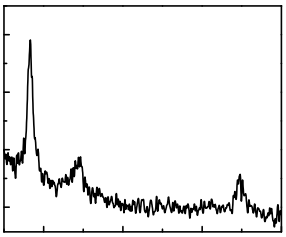


Fig.-5: XRD Pattern of Gold Nanoparticles

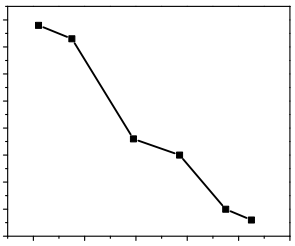


Fig.-6: Zeta Potential Values of Au NPs

Higher metal precursor concentration, greater the chances of obtaining AuNPs.[15] In present study the optimized concentration was found to be 0.5mM of Hydro chloro auric acid for 1ml of aqueous plant extract, as at higher concentration (1mM) agglomeration of nanoparticles was observed. According to previous studies, the pH of reduction reaction also affects the nanoparticle synthesis. More stable and smaller-sized AuNPs are produced at higher pH due to availability of biochemicals for reduction and stabilization of AuNPs. Moreover, the acidic pH prevents the generation of gold nanoparticles and thus produce unstable nanoparticles.[16] The enhanced stability of gold sol at higher pH might be due to growth of smaller gold nanoparticles over the generation of new gold nanoparticles from metal ions.[16,17] Similar results were obtained earlier in literature for the biosynthesis of gold sols using lemon grass and *Dioscorea bulbifera* tubers extract. It is pertinent to mention that *Akashmoni* contains saponins[18] their soapy nature makes them good surfactants. These saponins might acts as a templates for the generation of non-spherical gold nanoparticles. The particles behaviour at pH 5.9 might be due to the presence of various bio-molecules in extract which may be unable to stabilize particles at this pH. The TEM image (Fig. 4**)** shows that at pH 2.2 (inherent) the particles are polydispersed and show the presence of particles with anisotropic morphology like triangles, hexagons of average size of 30 nm. The average particle size of spherical shape nanoparticles was observed to be 8 nm. However, at pH 10.5 polygonal and nearly same size (20 nm) particles were obtained. The results are comparable with findings in literature for silver nanoparticles prepared with *Matoa* (Pometia pinnata) plant Extract. *C.* Imawan *et al.* found that at an alkaline condition, silver nanoparticles with small size and more regular shape were produced.[19] Further they found that at very high pH 11 and at low pH 5 formation of triangle and hexagonal silver nanoparticles occurs. However, zeta potential value for these nanoparticles is much lower (-16mv) than current work (-30 mv). Therefore, the method described in present work provides more stable sols of gold nanoparticles as compared to available methods present in literature.

Study Limitations and Future Directions

The presents work shows A green and quick way to synthesise gold nanoparticles (Au NPs) using the extract of *Acacia auriculiformis* (Akashmoni) without external reducing agents. The characterization was performed through Absorbtion spectroscopy (UV), Transmission Electron Microscopy, XRD, and zeta potential. However, this study has several limitations, including the lack of experimental work on the biological or catalytic applications of the Au NPs, phytochemical composition of *Akashmoni* extract in water to identify the active compounds responsible for nanoparticle reduction. In addition, further study is required on scalability of the method, reproducibility and surface functionalization of the synthesized gold nanoparticles for application such as drug delivery and sensing. Future research has scope in analysing the bioactivity and possible applications of these Au NPs.

Photochemical analysis maybe performed to know various chemical bio-molecules present in the extract to predict the detailed mechanism of gold ion reduction. Assessing toxicity and biocompatibility will also be helpful for sustainable application of current method for synthesis of AuNPs. It is also recommended to explore and compare the use all parts of *Akashmoni* in the preparation of gold as well as other metal nanoparticles.

CONCLUSION

A simple eco-friendly protocol has been developed for the preparation of monodispersed gold nanoparticles using Akashmoni pods extract used as both reducing and stabilising agent. The results of study reveal that the functional groups of biomolecules present in extract act as of stabilizing agent there by decreasing the interaction between the particles and preventing their aggregation. Therefore, the present work demonstrates that small and stable nanoparticles can be synthesized using essential stabilising properties of the extract. The observed pH effects on the shape and stability of nanoparticles further confirm this.

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CONFLICT OF INTERESTS

The authors declare that there is no conflict of interest.

AUTHOR CONTRIBUTIONS

The present work is related to *SDG-12: Responsible Consumption and Production*. All the authors contributed significantly to this manuscript, participated in reviewing/editing and approved the final draft for publication. The research profile of the authors can be verified from their ORCID ids, given below:

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