

Mimusops elengi bark extract mediated green synthesis of gold nanoparticles and study of its catalytic activity

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Abstract The bark extract of *Mimusops elengi* is rich in different types of plant secondary metabolites such as flavonoids, tannins, triterpenoids and saponins. The present study shows the usefulness of the bark extract of *Mimusops elengi* for the green synthesis of gold nanoparticles in water at room temperature under very mild conditions. The synthesis of the gold nanoparticles was complete within a few minutes without any extra stabilizing or capping agents and the polyphenols present in the bark extract acted as both reducing as well as stabilizing agents. The synthesized colloidal gold nanoparticles were characterized by HRTEM, surface plasmon resonance spectroscopy and X-ray diffraction studies. The synthesized gold nanoparticles have been used as an efficient catalyst for the reduction of 3-nitrophenol and 4-nitrophenol to their corresponding aminophenols in water at room temperature.

Keywords Gold nanoparticle · Green synthesis · *Mimusops elengi* · Polyphenols · Catalytic reduction · Nitrophenol

Introduction

Colloidal gold nanoparticles (AuNPs) has been the subject of tremendous research interest in recent years because of their applications in diversified areas such as catalysis, biotechnology, drug delivery, medicine and electronics. (Alkilany et al. 2013; Zhang et al. 2012; Murphy et al. 2008; Prati and Villa 2014; Thomas and Kamat 2003). Due to the large surface to volume ratio, the AuNPs have different physicochemical properties compared to the bulk solids. AuNPs exhibit different colors depending upon their size, shape and degree of aggregation (Weisbecker et al. 1996; Fujiwara et al. 1999; Aslan and Perez-Luna 2002; Mie 1908). The colloidal AuNPs stabilized with non-toxic biomolecules in aqueous medium are required for many of its applications. Among various synthetic methods, plant extract mediated solution phase synthesis of AuNPs involving reduction of Au(III) to Au(0) by plant extracts has gained profound significance in recent years because of the renewable and biocompatible nature of the plant extracts, eco-friendly aqueous medium and mild reaction conditions (Anastas and Kirchoff 2002; Irvani 2011). Moreover, since the plant extract itself acts as a stabilizer, it makes the method more advantageous over other synthetic methods. The extracts of *Acacia nilotica* leaf (Majumdar and Bag 2013), *Lantana camara* leaf (Dash et al. 2014a), *Abroma augusta* Linn bark (Das et al. 2014), *Breynia rhamnoides* (Gangula et al. 2011), *Saraca indica* (Dash et al. 2014b), *Piper betle* (Punuri et al. 2012), green coconut shell (Paul et al. 2014), etc., have been utilized for the synthesis of AuNPs. During our investigations on the utilization of triterpenoids (C30s) as renewable functional nano-entities (Bag et al. 2012; Bag and Dash 2011; Bag and Majumdar 2012; Bag and Paul 2012; Bag and Majumdar 2014), it occurred to us that the medicinally important bark extract

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of *Mimusops elengi*, rich in polyphenolic compounds, can be utilized for the synthesis AuNPs from HAuCl_4 (Baliga et al. 2011; Kadam et al. 2012).

Mimusops elengi (commonly known as Bakul) has been mentioned in all ancient scriptures of Ayurveda and has been used for medicinal purpose since centuries. The stem bark is dark gray in color, with striations and few cracks on the surface, internally reddish brown color having a bitter taste. The bark is used as a tonic, febrifuge, astringent, gargle for odontopathy, ulitis and ulemorrhagia (Baliga et al. 2011; Kadam et al. 2012). In the present study, the AuNPs have been synthesized using the bark extract of *Mimusops elengi* as a bio-reducing agent. The synthesized colloidal AuNPs were characterized by surface plasmon resonance spectroscopy, high-resolution transmission electron microscopy (HRTEM) and X-ray diffraction analysis.

Aminophenols are important precursors for the preparation of several analgesic and antipyretic drugs such as paracetamol, acetanilide and phenacetin. (Travis 2007). These compounds are also used as corrosion inhibitor in paints, anticorrosion lubricating agent in fuels, as photographic developer and in dye industries. Direct catalytic reduction of nitrophenols to aminophenols is of great significance to meet the current demand. In the present study, the catalytic activities of biosynthesized colloidal AuNPs have been investigated for the reduction of 3-nitrophenol (3-NP) and 4-nitrophenol (4-NP) in aqueous medium in the presence of sodium borohydride (NaBH_4) as a reducing agent. The kinetics of the reduction reactions investigated spectrophotometrically demonstrated that the colloidal AuNPs synthesized with lower concentration of plant extract were more efficient catalyst than that with higher concentration of plant extract.

Materials and methods

Materials

Au(III) solution

HAuCl_4 was purchased from SRL (Sisco Research Laboratory) and used without further purification. HAuCl_4 (35.4 mg) was dissolved in distilled water (10 mL) to obtain a 10.42 mM Au(III) stock solution. 4-Nitrophenol and 3-nitrophenol were purchased from MERCK and LOBA Chemie, respectively.

Preparation of the Mimusops elengi bark extract

The bark of *Mimusops elengi* was collected from the Vidyasagar University campus. Dried and finely powdered

Mimusops elengi bark (50.0 g) was suspended in ethanol (250 mL) and stirred magnetically at room temperature for 1 h and then filtered. Volatiles of the brownish filtrate were removed under reduced pressure to afford a brown crystalline solid (5.54 g). The crude material (0.660 g) was purified by column chromatography (si-gel, 100–200 mesh) using 30 % ethanol-ethyl acetate as the eluant affording a brownish shiny crystalline solid (0.550 g). The column purified bark extract (0.0504 g) was suspended in distilled water (10 mL) and sonicated in an ultra sonicator bath for 10 min to get a semi-transparent solution (5.04 g L^{-1}).

Synthesis of nanoparticles

Aliquots of Au(III) solution (0.16 mL, 10.42 mM each) were added drop-wise to the bark extract solution to prepare a series of stabilized AuNPs where concentration of the bark extract varied from 50 to 600 mg L^{-1} and the concentration of Au(III) was fixed at 0.42 mM. UV–Vis spectroscopic measurements of the solutions were carried out after 3 h of HAuCl_4 and *Mimusops elengi* bark extract was mixed.

Characterization

TEM images of AuNPs were recorded in JEOL JEM-2100 instrument. X-ray diffraction (XRD) analysis of the stabilized AuNPs was carried out in Rigaku Miniflex II diffractometer with Cu- $K\alpha$ radiation ($\lambda = 1.54 \text{ \AA}$). Mass spectra of the bark extract of *Mimusops elengi* were recorded in Shimadzu GCMS QP 2100 Plus. All the UV–Vis spectra were recorded in Shimadzu 1600 spectrophotometer.

Results and discussion

The presence of different types of plant secondary metabolites such as flavonoids, tannins, triterpenoids and saponins in the bark extract of *Mimusops elengi* has been reported (Baliga et al. 2011; Kadam et al. 2012; Rao et al. 2011). Indeed, mass spectral analysis of the bark extract of *Mimusops elengi* carried out in our laboratory indicated the presence of several polyphenolic compounds, steroids, etc. (supporting information Figure S1). Evidence for the presence of phenolic compounds in the bark extract of *Mimusops elengi* was also obtained from a positive ferric chloride test. The polyphenolic compounds present in the bark extract of *Mimusops elengi* can reduce Au(III) having a high reduction potential to Au(0) with concomitant oxidation of the polyphenolic compounds to the corresponding quinones. Collision of the Au(0) atoms with each

other can yield gold nanoparticles (AuNPs) and the AuNPs can be stabilized by the resulting quinone derivatives and other coordinating ligands present in the bark extract. The stabilizing ligands present on the surface of the AuNPs will hinder further aggregation of the AuNPs. To test this, we treated increasing concentrations of the aqueous bark extract of *Mimusops elengi* contained in vials with HAuCl₄ solution (Fig. 1). Visual observation of the appearance of pinkish red color almost instantly indicated the formation of AuNPs. On standing the vials at room temperature for several hours, intensification of the color was observed and then the intensities of the colloidal suspensions remained constant for several months indicating the stabilities of the synthesized AuNPs.

UV–visible spectroscopy studies

Due to charge transfer interactions between metal and the chloro ligands of HAuCl₄, the UV–visible spectrum shows two peaks at 222 and 293 nm (Fig. 1). On reaction with increasing concentration of the *Mimusops elengi* bark extract, disappearance of these two peaks was observed with concomitant appearance of a surface plasmon resonance (SPR) band between 536 and 541 nm region. Variation of color of the AuNP colloids has been reported arising due to the change in size, shape, composition, crystallinity, etc. (Alkilany et al. 2013). In our present studies, the SPR band was observed from 536 to 541 nm region and the intensities increased with increasing concentration of the bark extract from 50 mg L⁻¹ concentration to 600 mg L⁻¹ concentration. The strong peaks around 274–279 nm

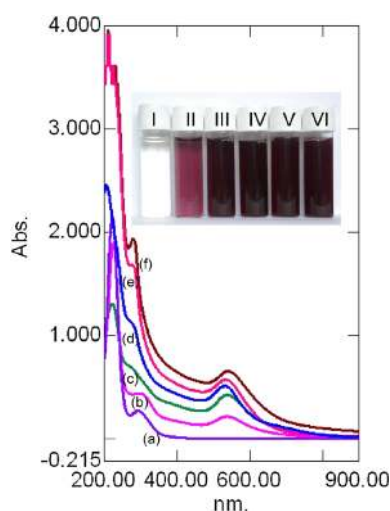


Fig. 1 UV–visible spectra of: *a* HAuCl₄ (0.42 mM), *b–f* AuNPs at 50, 100, 200, 400, 600 mg L⁻¹ concentrations of *Mimusops elengi* bark extract, respectively. *Inset* photograph of the vials containing *I* HAuCl₄ (0.42 mM), *II–VI* colloidal AuNPs stabilized by 50, 100, 200, 400, 600 mg L⁻¹ concentrations of *Mimusops elengi* bark extract, respectively (after one and half hour of mixing)

region at 400 and 600 mg L⁻¹ concentration are due to the formation of quinone moieties formed due to the oxidation of the phenolic compounds by Au(III).

HRTEM, EDX and XRD studies

To study the size distribution, shape and morphology of the stabilized AuNPs synthesized from the bark extract of *Mimusops elengi* at varied concentration, high-resolution transmission electron microscopy (HRTEM) was performed (Fig. 2; see supporting information Figure S2). Mostly spherical-shaped AuNPs of 9–14 nm size embedded in organic matrix in esthetically pleasing patterns were observed (Fig. 2j, l). The presence of the AuNPs held inside the organic matrix derived from the bark extract was clearly evident from Fig. 2d, e, g and h. Energy-dispersive X-ray analysis (EDX) was performed to determine the elemental composition of the AuNPs synthesized and stabilized by the *Mimusops elengi* bark extract. Strong peaks of Au in the area-profile analysis of the synthesized nanoparticles (Fig. 2k) confirm the formation of AuNPs. The presence of organic matrix was also confirmed from the characteristic carbon peak. For, X-ray diffraction analysis, the colloidal AuNP samples were coated over a glass plate and the volatiles were removed under 150 W electric bulb. After the removal of the volatiles, the X-ray diffraction analysis was performed. The characteristic reflections of the planes (111), (200), (220) and (311) of the face-centered cubic AuNPs at $2\theta = 38.24^\circ$, 44.45° , 64.71° and 77.78° , respectively (Figure S3) confirmed the formation of AuNPs. These values are in agreement with the reported standards JCPDS file no. 04-0784 for crystalline gold. The comparatively greater peak intensity of the (111) plane was observed which indicated the predominant orientation of the (111) plane.

Mechanism of the formation of stabilized AuNPs

The bark extract of *Mimusops elengi* is rich in different types of plant secondary metabolites such as flavonoids, tannins, triterpenoids and saponins. (Baliga et al. 2011; Kadam et al. 2012; Rao et al. 2011). Evidence for the presence of polyphenolic compounds was obtained from the ferric chloride test (supporting information). Mass spectral analysis of the bark extract carried out by us also supported the presence of the several polyphenolic compounds (Figure S1) including gallic acid (M⁺ 170), pinocembrin (M⁺ 256), quercetin (M⁺ 302), chlorogenic acid (M⁺ 354) or their analogs. A schematic representation of the possible mechanism for the formation of AuNPs and their stabilization by the polyphenolic compounds and other easily oxidizable compounds present in the bark

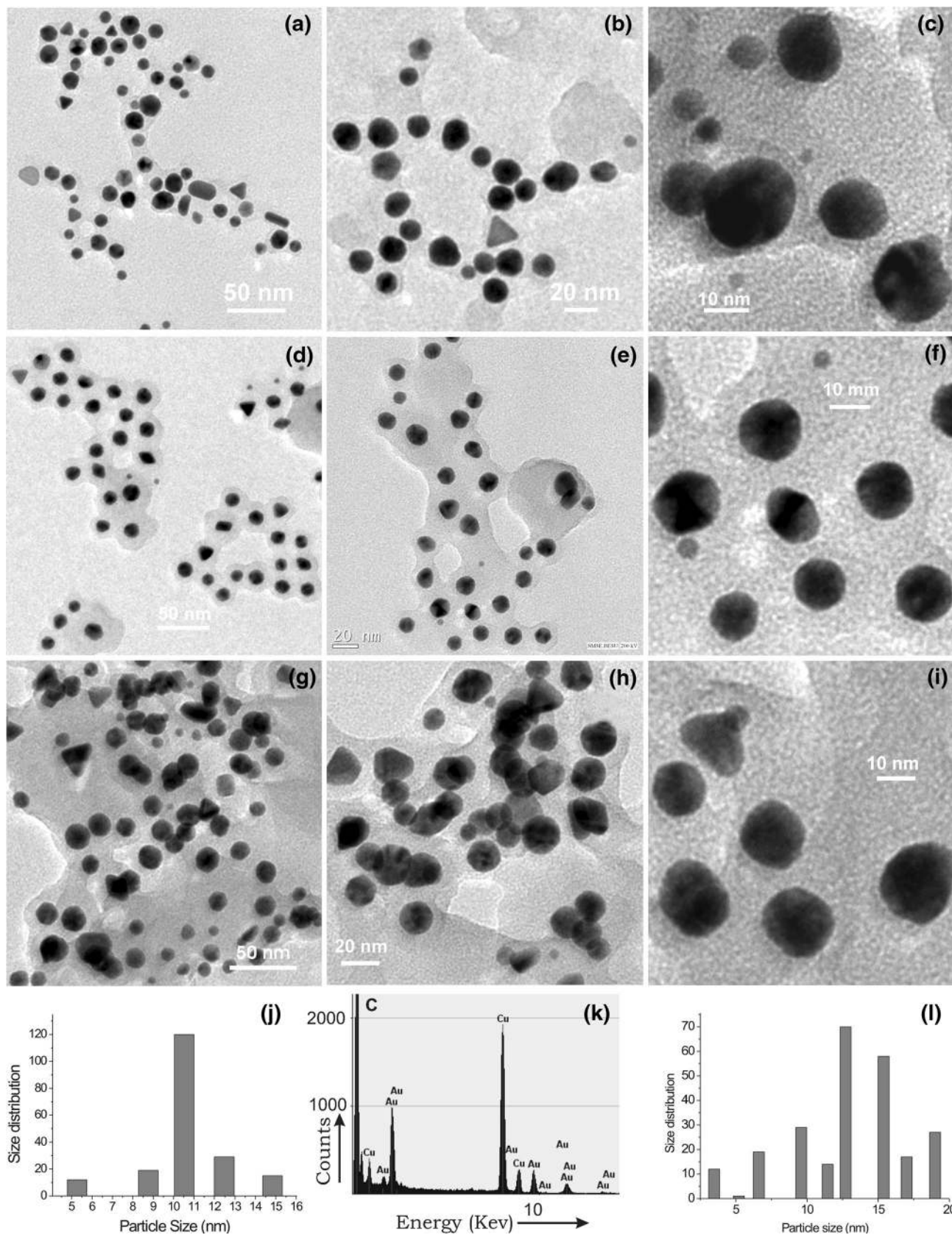


Fig. 2 a–c TEM images of AuNPs at 200 mg L⁻¹, d–f TEM images of AuNPs at 400 mg L⁻¹, g–i TEM images of AuNPs at 600 mg L⁻¹, j histograms of AuNPs at 200 mg L⁻¹, k EDX of AuNPs, l histogram of AuNPs at 400 mg L⁻¹

extract is shown in Fig. 3. Initially, ortho-dihydroxy compounds can form a five-membered chelate complex with Au(III). Then, the chelated Au(III) can be reduced to Au(0) with concomitant oxidation of the ortho-dihydroxy compounds to quinones. Collision of the neighboring Au(0) atoms with each other leads to the formation of the AuNPs which can be stabilized by the polyphenolic compounds, quinones as well as the other coordinating phytochemicals present in the bark extract (Huang et al. 2010).

Study of the catalytic activity of stabilized AuNPs

To study the catalytic activity of the synthesized colloidal gold nanoparticles using the bark extract of *Mimusops elengi*, two model reactions were carried out: (a) the reduction of 3-nitrophenol (0.05 mM) to 3-aminophenol and (b) 4-nitrophenol (0.05 mM) to 4-aminophenol by sodium borohydride (13–14 mM) at room temperature and the reactions were monitored by UV–visible spectroscopy (Fig. 4) (Wunder et al. 2011). The absorption band of 3-nitrophenol at 331 nm shifted to 394 nm in the presence of sodium borohydride due to the formation of 3-nitrophenolate ion. No transformation was observed on standing the mixture at room temperature for several days. This is due to a very large kinetic barrier for the reduction reaction. Interestingly, on addition of bark extract stabilized AuNPs (0.1 mL, synthesized with 100 mg L⁻¹ bark extract of *Mimusops elengi*) to the mixture, lowering of the intensity of the absorption peak at 394 nm was observed indicating the formation of 3-aminophenol. Complete disappearance of the 394 nm peak was observed within 300 s indicating completion of the reduction demonstrating the excellent catalytic activity of the synthesized colloidal AuNPs from the *Mimusops elengi* bark extract. The change in intensity of the absorption maxima of 3-nitrophenolate in alkaline medium (at 394 nm) with time provides a

reasonable tool for the study of its reduction kinetics. As the concentration of BH₄⁻ was much higher (approximately 300 fold) than that of 3-nitrophenol, its concentration remained practically constant during the reduction reaction and the reaction can be assumed as a pseudo-first-order reaction. A good linear correlation between ln(A/A₀) vs time was obtained (supporting information Figure S4a) and the value of the catalytic rate constant (*k*) was calculated to be 7.33 × 10⁻³ s⁻¹. Interestingly, on addition of double amount of stabilized AuNPs (0.2 mL, synthesized with 100 mg L⁻¹ bark extract of *Mimusops elengi*) to the mixture, the rate of the reduction reaction became much faster and the 394 nm peak completely disappeared within 30 s with concomitant formation of 3-aminophenol. Due to the very fast reaction, the catalytic rate constant could not be calculated for this reduction reaction. Increase in rates of the reduction reactions with increasing volume of the catalyst is due to the presence of larger number of reaction sites. Moreover, the rate of reduction reactions also became much faster when we used the AuNPs (0.2/0.1 mL) stabilized with 50 mg L⁻¹ bark extract of *Mimusops elengi* (Table 1) and the reduction was complete within 30 s.

Similarly, the absorption band of 4-nitrophenol at 319 nm was shifted to 401 nm after the addition of sodium borohydride due to the formation of 4-nitrophenolate ion. Here, also no further transformation was occurred on keeping the reaction mixture at room temperature due to the very large kinetic barrier of the reduction. Interestingly, on addition of bark extract stabilized AuNPs (0.2 mL, synthesized with 50 mg L⁻¹ bark extract of *Mimusops elengi*) to the mixture, lowering of the intensity of the absorption peak at 401 nm was observed indicating the formation of 4-aminophenol. Complete disappearance of the 401 nm peak was observed within 480 s indicating completion of the reduction demonstrating the excellent catalytic activity of the

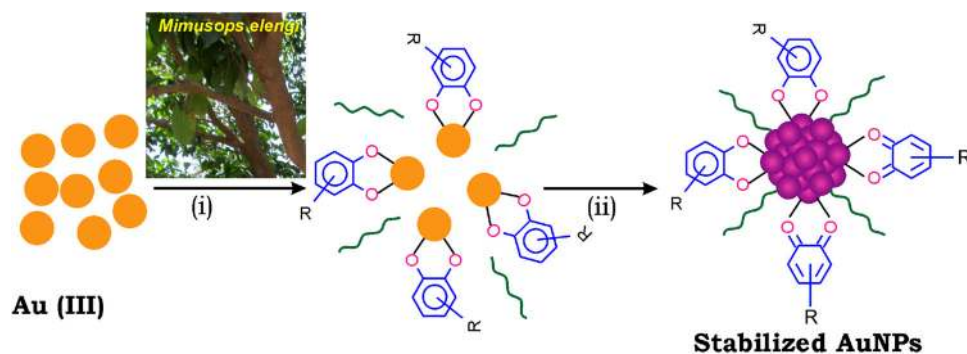


Fig. 3 Mechanism of formation and stabilization of gold nanoparticles by polyphenolic compounds and other plant metabolites (shown by green curved line) present in the bark extract of *Mimusops elengi*: *i* five-membered chelate ring formation by the polyphenolic

compounds of bark extract with Au(III), *ii* auto-reduction and stabilization by the polyphenolic compounds, quinones and other coordinating plant metabolites

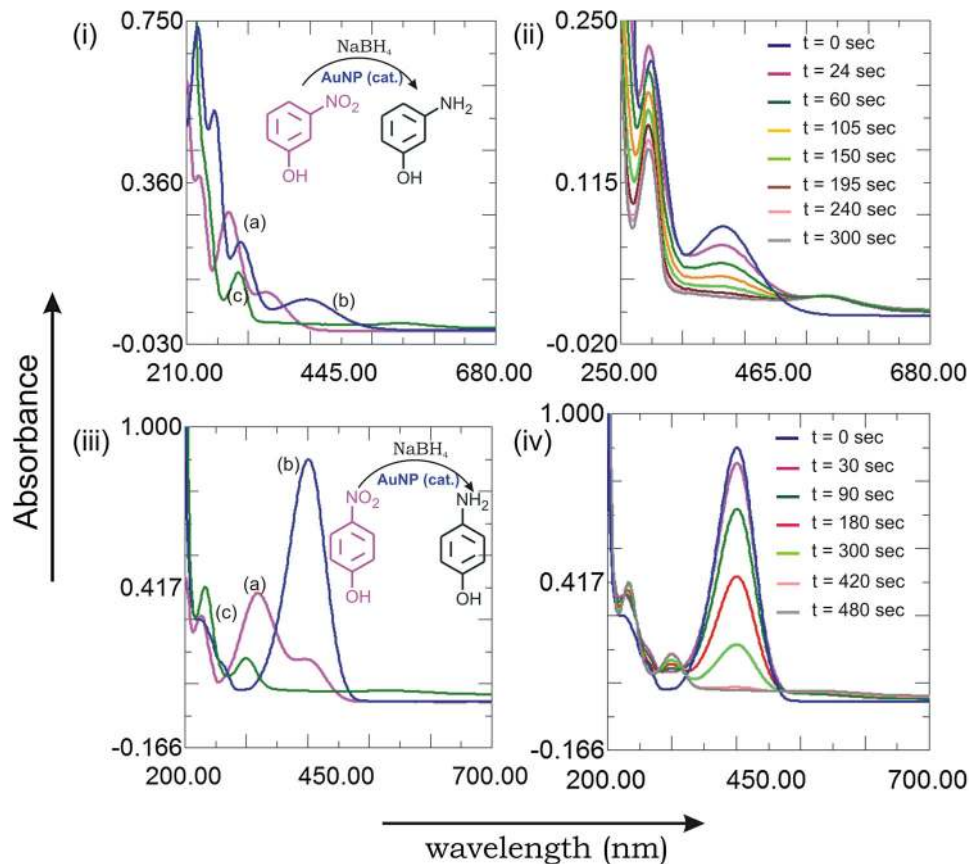


Fig. 4 **i, ii** UV–visible spectra during the catalytic reduction of 3-nitrophenol to 3-aminophenol using stabilized AuNPs (synthesized with 100 mg L^{-1} bark extract): **i** UV–visible spectra of **a** aqueous 3-nitrophenol solution (0.05 mM), **b** aqueous 3-nitrophenol solution (0.05 mM) in the presence of added sodium borohydride (15 mM), **c** after complete reduction of 3-nitrophenol using 0.1 ml of colloidal AuNPs as catalyst. **ii** Catalytic reduction of 3-nitrophenolate to 3-aminophenol at different time intervals using 0.1 ml stabilized AuNPs; **iii, iv** UV–visible spectra during the catalytic reduction of

4-nitrophenol to 4-aminophenol using stabilized AuNPs (synthesized with 50 mg L^{-1} bark extract): **iii** UV–visible spectra of **a** aqueous 4-nitrophenol solution (0.05 mM), **b** aqueous 4-nitrophenol solution (0.05 mM) in the presence of added sodium borohydride (15 mM), **c** after complete reduction of 4-nitrophenol using 0.2 ml of colloidal AuNPs as catalyst. **iv** Catalytic reduction of 4-nitrophenolate to 4-aminophenol at different time intervals using 0.2 ml stabilized AuNPs

Table 1 Catalytic activity of the *Mimusops elengi* bark extract stabilized AuNPs in the reduction of 3- and 4-nitrophenol by sodium borohydride

Entry	Substrate (mM)	NaBH ₄ (mM)	Colloidal AuNPs used		Reaction completion time (s)	Catalytic rate constant (k) (s^{-1})
			Volume (mL)	Conc. of bark extract used (mg L^{-1})		
1	3-NP/0.05	13.5	0.2	50	<30	Can't be calculated
2	3-NP/0.05	13.9	0.1	50	<30	Can't be calculated
3	3-NP/0.05	13.5	0.2	100	<30	Can't be calculated
4	3-NP/0.05	13.9	0.1	100	300	7.33×10^{-3}
5	4-NP/0.05	13.5	0.2	50	480	6.53×10^{-3}
6	4-NP/0.05	13.5	0.2	100	No reduction until 600 s	

synthesized colloidal AuNPs from the *Mimusops elengi* bark extract. In this case also, a good linear correlation between $\ln(A_t/A_0)$ vs time was obtained (supporting information Figure S4b) and the value of the catalytic rate

constant (k) was calculated to be $6.53 \times 10^{-3} \text{ s}^{-1}$. The observed rate constants were similar to that observed by us previously and others on similar systems (Majumdar and Bag 2013; Gangula et al. 2011).

During the course of the reactions, it was observed that the colloidal AuNPs synthesized and stabilized with 50 mg L⁻¹ bark extract of *Mimusops elengi* were more efficient catalyst than the colloidal AuNPs synthesized and stabilized with 100 mg L⁻¹ bark extract of *Mimusops elengi*. In the case of 3-nitrophenol reduction, with 50 mg L⁻¹ bark extract stabilized AuNPs (0.1 mL), the complete reduction occurred within 30 s whereas with 100 mg L⁻¹ bark extract stabilized AuNPs (0.1 mL), the complete reduction occurred within 300 s. Similarly, for 4-nitrophenol reduction, with the 50 mg L⁻¹ bark extract stabilized AuNPs (0.2 mL), the complete reduction occurred within 480 s but with the 100 mg L⁻¹ bark extract stabilized AuNPs (0.2 mL), no reduction was observed even after 10 min.

It is believed that, both 3-nitrophenol/4-nitrophenol and the reducing agent BH₄⁻ are adsorbed on the surface of the AuNPs and the surface hydride ions are then transferred to 3-nitrophenol/4-nitrophenol thereby facilitating the reduction reaction (Wunder et al. 2011). In both reductions of 3-nitrophenol and 4-nitrophenol, the rates were slower with AuNPs synthesized with 100 mg L⁻¹ concentrations of plant extract than that with 50 mg L⁻¹ concentration. This may be due to the fact that large excess of the plant extracts surrounding the AuNPs may sterically hinder the AuNPs for catalytic activity.

Conclusions

A very mild and efficient method for the green synthesis of colloidal gold nanoparticles has been demonstrated using the bark extract of medicinally important *Mimusops elengi*. According to our knowledge, this is the first report of the synthesis of AuNPs using *Mimusops elengi* bark extract. The polyphenolic compounds present in the bark extract acts as an effective reducing agent as well as stabilizing agent and AuNPs of 9–14 nm size were formed. The application of the synthesized colloidal AuNPs has been demonstrated using it as efficient catalyst for the reduction of 3-nitrophenol to 3-aminophenol as well as 4-nitrophenol to 4-aminophenol at room temperature in aqueous medium. As the aminophenols are important precursors for the preparation of many industrially useful compounds, direct catalytic reduction of nitrophenols to aminophenols described here has tremendous significance for its industrial applications.

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