ORIGINAL ARTICLE

# Green synthesis and characterization of iron nanoparticles from *Bauhinia tomentosa*

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## Abstract

**Introduction:** Synthesis of metallic nanoparticles using plant extract is inexpensive, easily scaled up, and eco-friendly and their technique has wide open applications in various sectors such as medicine, health, and environment due to its mechanism of action for the betterment of human. The aforesaid challenges and principles of nanoparticles have advanced in the green synthesis utilizing the phytoconstituents to reduce and stabilize the metallic nanoparticles. **Materials and Methods:** In the investigation, green synthesis of iron nanoparticles from *Bauhinia tomentosa* was characterized under ultraviolet–visible spectrophotometer, Fourier transform infrared at various concentrations and durations. The characterized iron nanoparticles were analyzed for reducing assay and hydrogen peroxide assay to prove their antioxidant properties and antibacterial properties. **Results and Discussion:** When the plant extract was added to ferric chloride solution $Fe^{3+}$ reduced to $Fe^{0}$ which was indicated by the color change from green into blackish green proved the formation of iron nanomaterials (due to surface Plasmon resonance singularity). The synthesized iron nanomaterials were characterized by different parameters such as proportions (extract: Ferric chloride) and development study at different durations which are identified as factors affecting the yields of nanoparticles and later the nanoparticles were characterized. The reducing power of the iron nanoparticles synthesized from *B. tomentosa* was investigated by comparing with the reductive ability of ascorbic acid. Reducing power assay has proved to be one of the convenient and rapid screening method for measuring the antioxidant potential. **Conclusion:** In the present investigation, the plant bioactive constituents of *B. tomentosa* proved to be highly efficient as reducing agents, antibacterial agent which on further clinical investigation will prove to have high therapeutic value in the medical field.

**Key words:** Antibacterial, Antioxidant, Green synthesis, Phytoconstituents, Resistant, Sensitive, Therapeutic value

## INTRODUCTION

Biosynthesis of nanoparticles is considered to be significant in modern techniques in the field of material science, life sciences, etc., due to its physicochemical properties such as extreme small size, large surface area to volume ratio, biocides, and antimicrobial.[1] Advancement in nanoparticle synthesis with improved production of energy, change in thermophysical properties, implementation in modern devices/technology etc. will improve the quality of materials and matter and utilization in various industrial sectors.[2-7] Physical nature of nanoparticles as solid (either amorphous or crystalline),[8] higher surface to volume ratio, catalytic reactive, etc., increased their surface energy in the biological studies[9,10] where they are able to adsorb or encapsulate the bioactive compounds or drug or phytoconstituents and has high enzymatic degradation.[11] Traditional herbal medicines are used in almost developed and developing countries due to its cultural and spiritual perspectives and provide a good alternative in the pharmacological activities.[12,13] Phytoconstituents play an important role in the green synthesis of nanoparticles due to its biocompatibility and cost effective and it does not involve any external force (high energy, temperature, pressure, etc.).[14-19] Evidence proved the production of silver and gold nanoparticles from *Datura metel*[20,21] by sunlight exposure methods.

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In the present investigation, iron nanoparticles were synthesized from *Bauhinia tomentosa* with confidence that iron nanoparticles are employed in the treatment of water since 1990 due to its unique physicochemical properties such as adsorption, large reactive and specific surface area for degradation,[22,23] and relatively expensive.[24] Research evidences of iron nanoparticle synthesis from various plants like *Terminalia chebula* fruit,[25] banana peel,[26] tea leaf,[27,28] *Eucalyptus*[29] and *Trdax procumbes*[30] were investigated and recorded. *B. tomentosa* commonly known as the yellow bell orchid tree belongs to the Fabaceae family which comprises more than 300 species and is found in tropical areas. The word "tomentosa" means hairy and it refers to the velvety/hair pods.[31] The genus *Bauhinia* is well-known plant in herbal medicine in the therapeutic efficacy of its different species among which the species *tomentosa* is commonly known as "Kanjana" in Tamil and "Phalgu" in Sanskrit, and also stated that the dried leaves, buds, and flowers are prescribed for dysentery.[32] Iron nanoparticles were synthesized and characterized from *Sorghum* Moench[33] and *Euphorbia milii.*[34]

## MATERIALS AND METHODS

Disease-free fresh plants (*Bauhinia tomentosa*) were collected and cleaned thoroughly with continuous tap water to remove the dust and sand particles. Twenty grams of spliced leaves were weighed and ground with mortar and pestle by adding demineralized water and to it, 1 mM of aqueous ferric chloride solution was added. The mixture was incubated for 20–30 min and the color change was observed from dark green into bluish-green which indicated the synthesis of iron nanoparticles.

### Characterization Studies

The synthesis of iron nanoparticles at different time duration was confirmed using ultraviolet (UV)–visible spectrophotometer of ELICO SL 171 Mini Spec from the range of 400 nm–700 nm. The graph was plotted between absorbance and wavelength and further the dried iron nanoparticles were analyzed using Fourier transform infrared (FT-IR) with spectral range of 500–4000 $cm^{-1}$.

### Antioxidant Activity Studies

a. Reducing power assay

Iron nanoparticles of different concentrations such as 20, 40, 60, 80, and 100 were prepared and mixed with 2.5 mL of phosphate buffer (pH 6.6) and 2.5 mL of 1% of potassium ferricyanide solution and the mixture was incubated in water bath at 50°C for 20 min. The reaction was stopped by adding 2.5 mL of 10% trichloroacetic acid and then solution mixture was centrifuged at 3000 rpm for 10 min. The upper layer was transferred into sterile test tube and mixed with distilled water and ferric chloride solution. The absorbance at 700 nm was measured as the reducing power using UV spectrophotometer.

b. Hydrogen peroxide scavenging activity

Iron nanoparticles of different concentration were prepared and hydrogen peroxide solution was prepared with phosphate buffer solution and incubated for 10 min. The absorbance was measured at 560 nm using a UV spectrophotometer against hydrogen peroxide blank solution.

The percentage of hydrogen peroxide scavenging by the iron nanoparticles was calculated using the formula:

$$\%\ \text{Scavenging} = \frac{\text{Absorbance control} - \text{Absorbance sample}}{\text{Absorbance control}} \times 100$$

### Antimicrobial activity studies

Pure cultures of *Escherichia coli*, *Salmonella* Typhi, and *Staphylococcus aureus* were obtained from a microbial type culture collection, Institute of Microbial Technology, Chandigarh and were cultured in nutrient broth for antibacterial study (well diffusion method) in the present study. The test organisms were swabbed onto the duplicate petri plates and four wells were made with the help of sterile cork borer. Different concentrations of iron nanoparticles were loaded into four wells in petri plate, respectively, and antibiotic kanamycin was used as positive control. Then, the plates were incubated for 24 h at optimum temperature. The zones of inhibitions (in mm diameter) were recorded after 24 h.

## RESULTS

Diseased free plant *B. tomentosa* was collected, cleaned in tap water and the leaf extract was subjected to produce iron nanoparticles. When the plant extract was added to ferric chloride solution $Fe^{3+}$ reduced to $Fe^0$ which was indicated by the color change from green into blackish-green proved the formation of iron nanomaterials (due to surface Plasmon resonance singularity). The synthesized iron nanomaterials were characterized by different parameters such as proportions (extract: ferric chloride) and development study at different durations which are identified as factors affecting the yields of nanoparticles and later the nanoparticles were characterized under UV–visible spectrophotometer in the range of wavelength from 400 to 700 nm. Synthesized iron nanoparticles were analyzed by UV spectra at different proportions (1:1, 2:2, 3:3, 4:4, and 5:5) and the development of nanoparticles recorded a significant size with the increase in the proportions and the results were plotted in the graphs (Figures 1-5). The synthesized iron nanoparticles were characterized at different durations such as 30 min, 1 h, 2 h, 3 h, and 4 h; the result showed maximum absorbance at

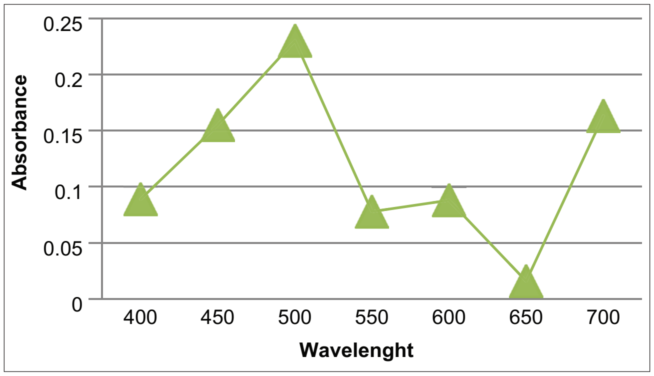


**Figure 1:** Iron nanoparticles development from *Bauhinia tomentosa* at 1:1 ratio

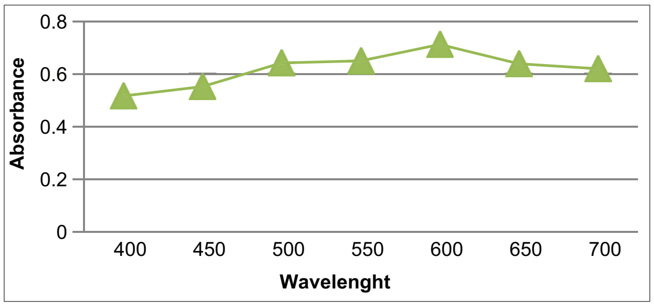


**Figure 2:** Iron nanoparticles development from *Bauhinia tomentosa* at 2:2 ratio

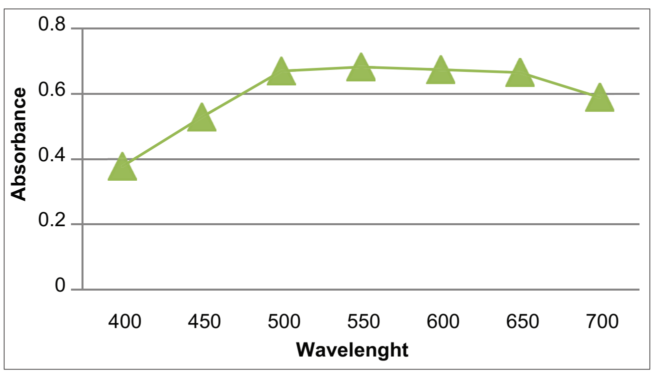


**Figure 3:** Iron nanoparticles development from *Bauhinia tomentosa* at 3:3 ratio

550 nm, with a gradual decrease in absorbance with respect to time and the absorbance was plotted graphically in Figure 6. The FT-IR spectra of synthesized iron nanoparticles showed vibration bands at 3265.49 $cm^{-1}$ which indicated alcohol and phenol group stretching and the peak at 2929.87 $cm^{-1}$ indicated alkyl C-H stretch. Aromatic C=C bonding bending was observed at 1602.85 $cm^{-1}$, C-H stretching was registered at 1394.53 $cm^{-}$1, C-H and C-O stretching at 1016.49 $cm^{-1}$, respectively (Figure 7).

The reducing power of the iron nanoparticles synthesized from *B. tomentosa* was investigated by comparing with the reductive ability of ascorbic acid. Reducing power assay has proved to be one of the convenient and rapid screening method for measuring the antioxidant potential. The reducing power of

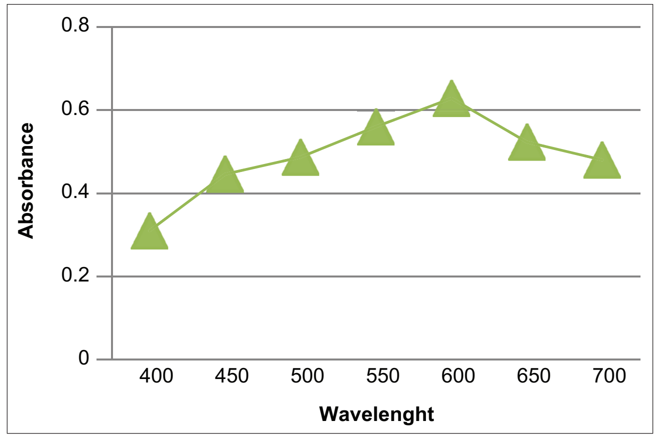


**Figure 4:** Iron nanoparticles development from *Bauhinia tomentosa* at 4:4 ratio

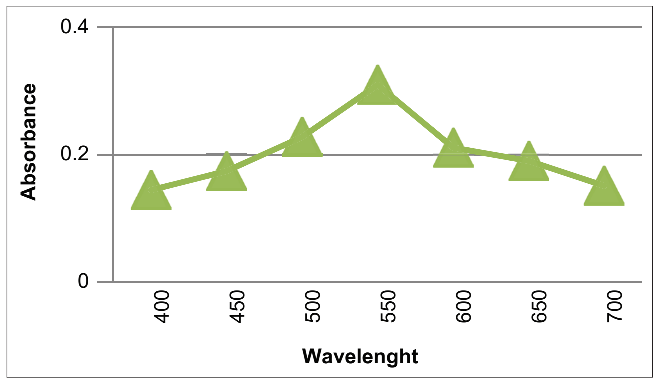


**Figure 5:** Iron nanoparticles development from *Bauhinia tomentosa* at 5:5 ratio

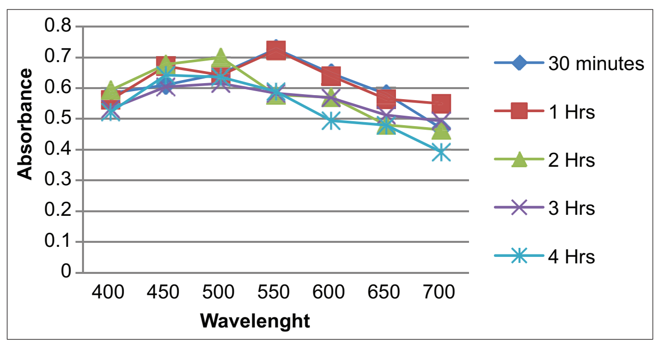


**Figure 6:** Characterization of synthesized iron nanoparticles by ultraviolet spectrophotometer analysis at different durations

the nanoparticles increased significantly with concentration as compared with ascorbic acid, as shown in Figure 8. Hydrogen peroxide scavenging activity of synthesized nanoparticles was investigated against ascorbic acid (Figure 9). Free radicals have the potential to oxidize proteins, lipids, and DNA in cells and inhibit the oxidative mechanisms that lead to degenerative diseases. Antioxidants play an important role as health protecting factors since they have the ability to trap such free radicals which are of plant derived compounds.

*B. tomentosa* plant is found to be wide spread and proved to have high therapeutic value in the medical field. The

**Table 1:** Antibacterial activities of synthesized iron nanoparticles of *Bauhinia tomentosa*

| S. No. | Microorganism | Iron nanoparticles | | | |
|---|---|---|---|---|---|
| | | Zone of inhibition in mm diameter | | | |
| | | 30 µL | 60 µL | 90 µL | 120 µL |
| 1. | *Escherichia coli* | 1.4 | 1.7 | 1.9 | 2.1 |
| 2. | *Staphylococcus aureus* | 0.5 | 1.5 | 1.8 | 2.0 |
| 3. | *Salmonella* Typhi | 1.0 | 1.4 | 1.7 | 2.0 |

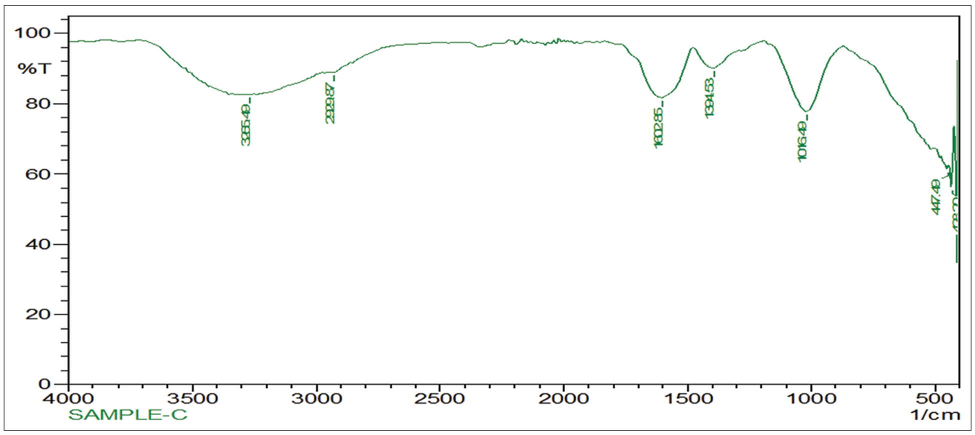


**Figure 7:** Fourier transform infrared result of iron nanoparticles of *Bauhinia tomentosa*

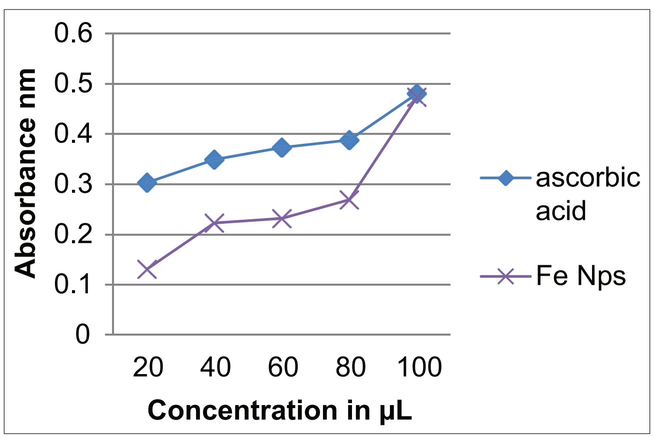


**Figure 8:** Antioxidant property of synthesized iron nanoparticles by reducing power assay method of *Bauhinia tomentosa*

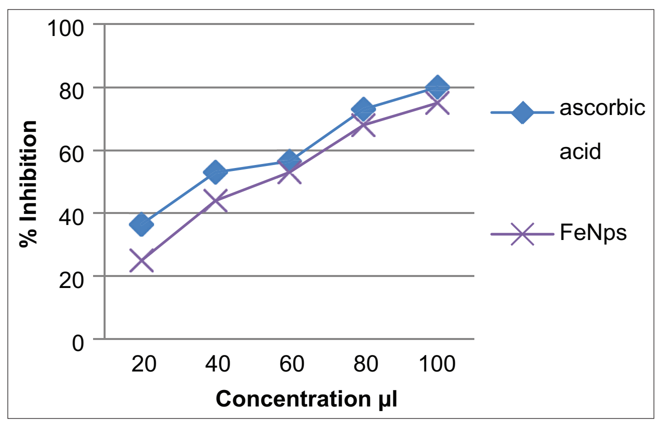


**Figure 9:** Antioxidant property of synthesized iron nanoparticles by hydrogen peroxide assay method of *Bauhinia tomentosa*

synthesized iron nanoparticles registered high zone of inhibition when it was subjected to antimicrobial activity study at different concentration 30 µL, 60 µL, 90 µL, and 120 µL. The iron nanoparticles of *B. tomentosa* were found to be highly sensitive to all the three microbes. The maximum zone of inhibition (2.1 mm) was observed in *E. coli* followed by *S. aureus* and *Salmonella* Typhi (Table 1).

## DISCUSSION

The maximum absorption peak was registered at 432 nm with leaf extract of *D. metel*[35] and it was proved to be due to the excitation of longitudinal plasma vibration of silver nanoparticles,[36-38] the size of the nanoparticles can even be estimated visually by the broadness of peak and it also proved that the increase of particle size, the peak becomes narrower with a decreased bandwidth.[39,40] Iron nanoparticles were synthesized based on the presence of polyphenols from *Mangifera indica*, *Murraya koenigii*, *Azadirachta indica*, and *Magnolia champaca* and the reduction of $FE^{2+}$ ions was confirmed and peaks were obtained between 300 nm and 500 nm.[22] The iron nanoparticle peaks were recorded differently for various extracts such as 216–256 nm;[41] 256–277 nm;[42] and 296–325 nm[43] under UV-VIS spectrophotometer.

C-O and C=O stretching vibrations were observed at 1205 and 1745 $cm^{-1}$, OH and sulfate groups at 3257 and 1095 $cm^{-1}$ and C-N stretching vibration for aromatic amines at 1367 $cm^{-1}$ during FTIR analysis of iron nanoparticles.[41] The wavelength band peak was recorded for Al-FeNPs where OH bond registered at 3597 and 3600 $cm^{-1}$ and C+O stretching vibration at 1688 and 1726 $cm^{-1}$.[44] Therefore, it is proved that the formation of nanoparticles is due to the presence of polyphenolic compounds which are responsible for direct reduction of iron ions to zero-valent particles.[45]

Free radicals have the potential to oxidize proteins, lipids, and DNA in cells and inhibit the oxidative mechanisms that lead to degenerative diseases. Antioxidants play an important role as health protecting factors since they have the ability to trap such free radicals which are of plant-derived compounds.[46] Molecules that are capable of preventing the oxidation of other molecules are known as antioxidants and these molecules are very important in the treatment of various diseases. Recently, work has been carried out to explore the antioxidant property of CuO nanoparticles and recorded free radical scavenging activity up to 85% in 1 h and it was comparatively higher than the other metallic nanoparticles.[47]

The antibacterial activities of silver nanoparticles at 100 µg/mL concentration were efficient in inhibiting *Pseudomonas aeruginosa* and *E. coli.*[48] The antibacterial activity in terms of zone of inhibition increased with an increase of stack solution from 50 to 200 µL and the mechanism associated is generation of reactive oxygen species, bonding of Ag+ ions with sulfhydryl groups[25] and penetration through cell wall and cause damage.[49] The Gram-positive bacteria were more sensitive to silver nanoparticles than Gram-negative bacteria (minimum concentration was 1.5 mg/mL for *S. aureus* and 6 mg/mL for *E. coli*).[50] The antimicrobial study of iron oxide nanoparticles synthesized by coprecipitation method showed high susceptible against *E. coli* when compare to *S. aureus* was reported.[51]

## CONCLUSION

The simple procedure involved to synthesis iron nanoparticles using *B. tomentosa* proved its efficiencies and effectiveness in all the analysis during the period of study. The phytoconstituents of *B. tomentosa* also mediated all the reduction process and also acted as a capping agent and thereby proved to have sensitivity against Gram-positive and Gram-negative bacteria.

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