

Synthesis and Characterization of Platinum Nanoparticles (Pt-NPs) from *Centella Asiatica L* Leaf Extract

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Abstract.

Nanotechnology is a burgeoning field and is widely applied to biomedical engineering and nanomedicine. Platinum nanoparticles (Pt-NPs) are noteworthy scientific tools that are being explored in various biotechnological, nanomedicinal, and pharmacological fields. They are unique because of their large surface area and their numerous catalytic applications such as their use in automotive catalytic converters and as petrochemical cracking catalysts. Pt-NPs have been widely utilized not only in the industry, but also in medicine and diagnostics. The whole world has been utilizing plants for basic preventive and curative health care since time immemorial. Several plants have been search by the human race for the control of diseases. Human beings have used plants for the treatment of diverse ailments for thousands of years. According to the World Health Organization, over 80% of the world's population, or 4.3 billion people, rely upon traditional plant-based systems of medicine to provide them with primary health care. In this study, the Platinum nanoparticles are synthesized from *Centella asiatica L* leaves and characterize the nanoparticles by UV-VIS, SEM, XRD. The UV-VIS profile showed the peaks at 297.55, 300.25, 346.11 and 467.21 nm with the absorption 2.2307, 2.1279, 1.8237 and 1.0017 respectively. The X-ray structural diffraction pattern of the Pt-NPs produced using *Centella asiatica L* was proved and confirmed by the characteristic peaks observed in the XRD images. The SEM images showed the aggregates of reduced platinum nanoparticles. The changing of colour from yellow to brown then black indicates the synthesis of platinum. The results of this study suggested that the Pt-NPs synthesis from *Centella asiatica L* leaf extracts have many pharmacological properties which may used in the development of new drugs for maintenance of good health

Keywords: *Centella asiatica L*, Pt-NPs, UV-VIS, XRD, SEM etc

INTRODUCTION:

Nanotechnology is the most promising field for generating new applications in medicine. However, only few nano-products are currently in use for medical purposes. The ability to manipulate the shape and size of materials at nano-scale has revolutionized the scientific world. Nano-sized materials have distinct physical, chemical, electrical and optical properties which have broadened their applications in various fields like catalysis, electronics, medicine and water purification¹. Consequently, it has triggered research interest towards the synthesis of noble metal nanoparticles via several chemical and biological methods.

The most prominent nanoproduct is PNPs. PNPs are generally smaller than 100 nm and contain 20 to 15,000 platinum atoms. Nanoparticles are often in the range 1 to 100 nm, and this is the size as that of human proteins. Metal nanoparticles possess a very high surface-to-volume ratio. In biology and biochemistry, nanoparticles have attracted much attention. Especially, PNPs with size in the range of 10 to 50 nm are most attractive for practical reasons²

Common biological methods for synthesis of NPs include several organisms such as bacteria, actinomycetes, algae, and fungi. Although microorganisms are exploited for the synthesis of Pt-NPs, controversy still exists regarding the use of microorganisms because the production time of NPs is high because of the time required to grow bacterial/fungus cultures and for bacterial cell maintenance. Therefore, researchers are interested in exploiting the use of plants and plant extracts, which are readily available and abundant and do not require any

media to grow. Plant-based synthesis of NPs has numerous advantages over the other types of biological methods³. The present study are focused to synthesize the Platinum nanoparticles from *Centella asiatica* L leaf extracts and elucidate its characterization.

METHOD:

Collection of Plant Material

Plant source selected for the present study was *Centella asiatica* L. Leaf part of the selected plant was collected from in and around Namakkal.

Preparation of Plant extract and Biosynthesis of Pt-Nps (Platinum Nanoparticles)⁴

Hexachloroplatinic acid (H₂PtCl₆.6H₂O) was purchased from Sigma - Aldrich and used without any purification. Fresh leaves of *Centella asiatica* L were collected and thoroughly washed. Deionised water was used throughout the experiments.

10 g of finely cut fresh leaves of *Centella asiatica* L was extracted with 200 mL of distilled water at 60°C for 10 min and filtered. The filtrates were stored at 4°C for further use. In a typical synthesis, 5 mL of the leaves extract was added to 50 mL aqueous solutions of hexachloroplatinic acid (1 x 10⁻³ M) in a sealed 3-necked flask and maintained at 90 °C for 1 h. Biosynthesized Pt-NPs was monitored visually and by using UV/Vis spectrophotometer in the range 200–700 nm

X-RAY Diffraction (XRD) Analysis⁵

X-ray diffraction (XRD) patterns were recorded on an X-ray diffractometer (PW1710, Philips), using Cu K α radiation ($\lambda = 1, 54060>$) at 40 kV and 30 mA. The diffraction angle ranged from 5 to 90°. The crystallite size of the Pt nanoparticles was calculated based on X-ray diffraction measurements. The crystallite size was calculated from the full width at half maximum (FWHM) of peak using the Scherrer formula.

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

where *L* is the average crystallite size of the Pt particles, *K* is a constant of 0.9, λ is the X-ray wavelength, β is the FWHM in radians, and θ is the diffraction angle.

Scanning Electron Microscopy⁶

The supernatant from the maximum time-point of production of Platinum nanoparticles was air-dried. The synthesized Platinum nanoparticles were fabricated on a glass substrates were done for the determination of the formation of Platinum nanoparticles. The morphology and size of Platinum nanoparticles was investigated using Scanning Electron Microscope (VEGA 3 TESCAN).The micrograph were recorded by focusing on clusters of particles.

UV-VISIBLE Spectral Analysis⁷

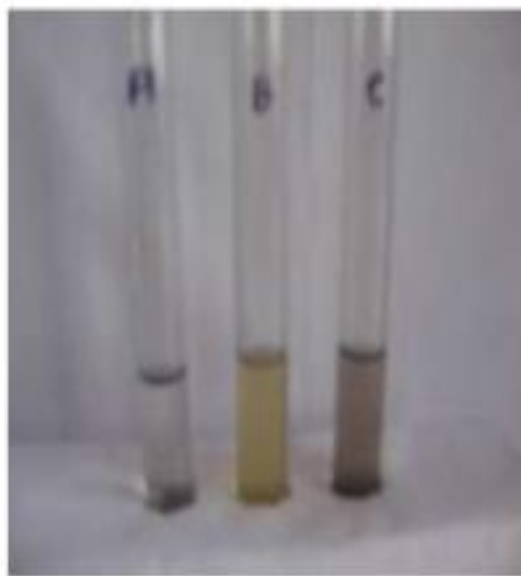
Switch on the instrument. Leave it for the instrument for 15 min machine calibration. Run the software by double clicking on the icon Double Beam Spectrophotometer present on the desktop. Switch on the Vis and UV lamps by mouse click on the yellow and blue icons from tool bar. It takes some time period (~ 3 minutes) for UV lamp to become ready .Fill the two cuvettes, one cuvette with appropriate background solution and the other with the sample. Place the two cuvettes in appropriate sample holders. After the blank solution was kept and autocorrect it to zero. Then kept sample to read the absorbance at specific ranging from 200-900 nm with the minor unit of 250.The values are tabulated and therefore the graph was plotted.

RESULTS:

Table No: 1: Indication of Colour Change in the Synthesis of Pt-NPs from *Centella asiatica* L Leaves extract

S.no	Plant leaf extract+ H ₂ PtCl ₆ Solution	Colour change		pH change		Colour intensity	Time	Result
	Scientific name	Before	After	Before	After			

1	<i>Centella asiatica</i> L	Light green	Brown	8.0	6.0	+++	20 min	Positive
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A- H_2PtCl_6 Solution B- *C.asiatica* L extract C- Pt-NPs
Fig. No: 1 Visual inspection of Colour Change in the Synthesis of Pt-NPs from *Centella asiatica* L Leaves extract

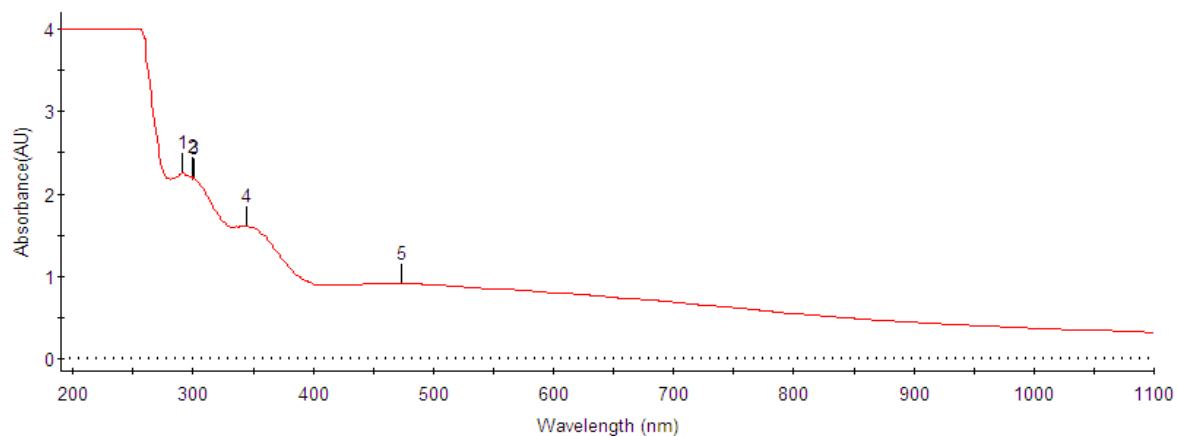


Fig. No: 2 UV-VIS analysis of Pt-NPs synthesis from *Centella asiatica* L leaf extract

Table No: 2 UV-VIS analysis of Pt-NPs synthesis from *Centella asiatica* L leaf extract

S.no	Wave Length (nm)	Absorbance (AU)
1	297.55	2.2307
2	300.25	2.1279
3	346.11	1.8237
4	467.21	1.0017

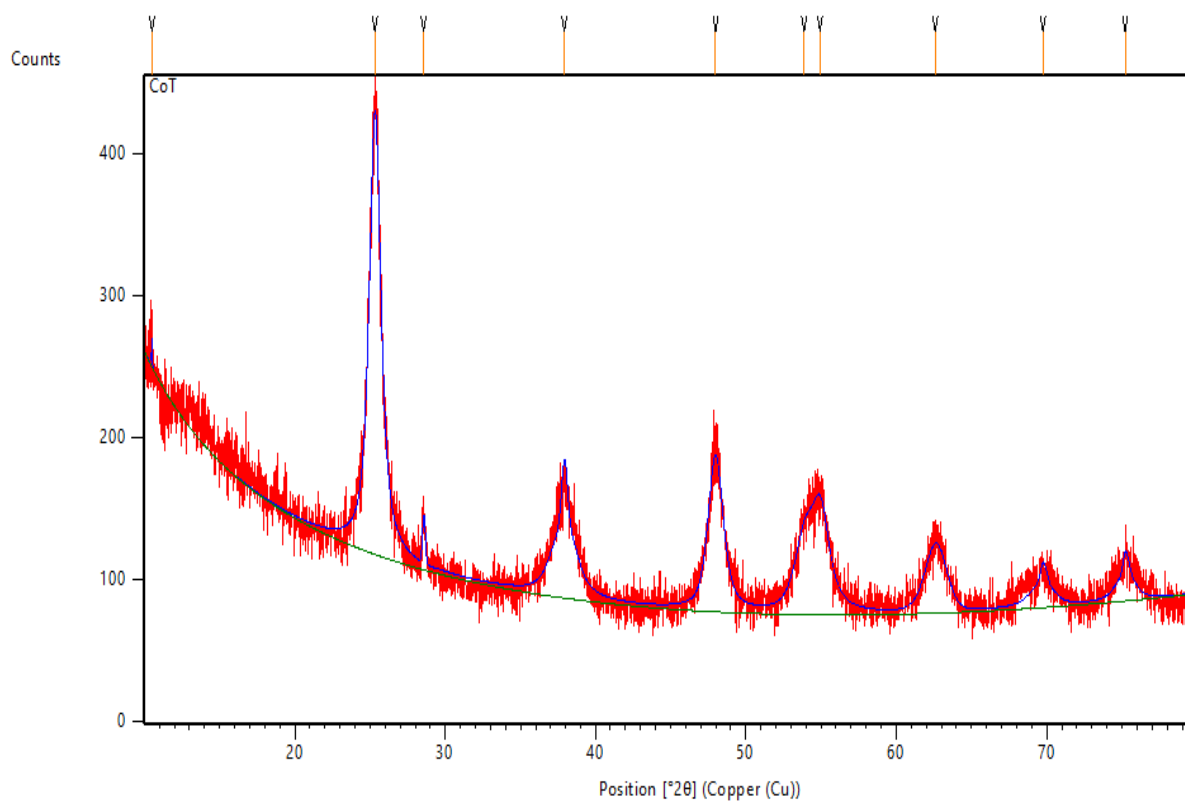


Fig. No: 3 XRD analysis of the reduced Platinum

Table No: 3 XRD analysis of the reduced Platinum

Pos. [°2θ]	Height [cts]	FWHM Left [°2θ]	d-spacing [Å]	Rel. Int. [%]
10.5118	25.74	0.0548	8.40896	12.31
25.3164	209.13	0.9211	3.51519	100.00
28.5518	24.42	0.1967	3.12379	11.68
37.9012	66.00	0.8568	2.37195	31.56
47.9582	73.69	1.1886	1.89540	35.24
53.8868	32.35	1.5972	1.70003	15.47
54.9410	44.05	1.3907	1.66987	21.06
62.5909	32.67	1.7180	1.48291	15.62
69.7670	22.51	0.9060	1.34689	10.76
75.2214	26.00	0.6400	1.26218	12.43

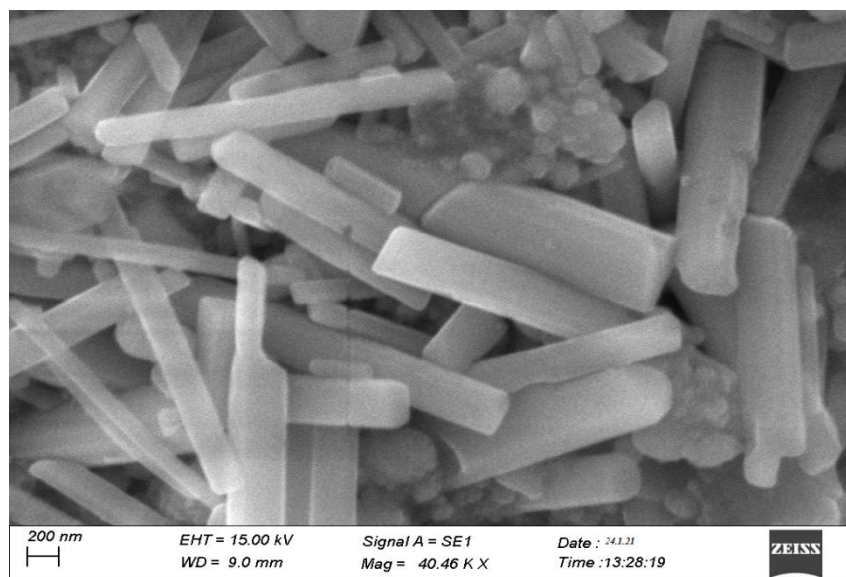


Fig. No: 4 SEM Micrograph of synthesized Pt-NPs from *Centella asiatica* L

The high temperature is required for platinum reduction rate faster. In this study, the temperature was maintained at 100 °C and the yield about 100% was recovered within an hour. It is well understood that 1 g of chloroplatinic acid generally contains about 0.4 g of platinum metal ions. The equal amount was recovered from plant extract are claimed as 100% recovery. Qualitative analysis of the colour change of the platinum (IV) solution from very light yellow to mild brown indicates the development of platinum (II) and the colour change from mild brown to black indicates the formation of platinum (0) (Table 1 & Fig. 4). The equal amount of platinum solution and plant leaf extract was maintained separately under the same reaction conditions for control experiments. The colour change was not noticed.

The rapid reduction of PtCl₂ with 30%, 70 %, 90 % after after 2 h, 4 h and 8 h using plant extract. The bioreduction of chloroplatinic acid with reaction temperature of 100 °C using tulsi leaf extract to reduce the reaction time with greater efficiency. The conversion of silver and gold was also noted within 11 and 3 min at 95° C using Magnolia leaf broth. The rate of platinum nanoparticle synthesis increased with increases in reaction temperature. At a reaction temperature of either 25 or 60 °C, 20% of platinum ions were converted to platinum nanoparticles⁸. The relatively less rate of platinum nanoparticle synthesis is possibly due to a difficulty in initially forming platinum nuclei, indicating that achieving close to 100% conversion to platinum nanoparticles requires longer reaction times and higher temperatures than those required for either gold or silver nanoparticles⁹

The qualitative UV-VIS profile of ethanolic extract of Pt-NPs synthesis from *Centella asiatica* L leaf was taken at the wavelength of 200 nm to 1100nm due to the sharpness of the peaks and proper baseline. The profile showed the peaks at 297.55, 300.25, 346.11 and 467.21 nm with the absorption 2.2307, 2.1279, 1.8237 and 1.0017 respectively (Table 3). Figure 7 shows the absorption spectrum of Pt-NPs and these are almost transparent in the wavelength region of 200-1100 nm

Absorption bands observed pertaining to Pt-NPs synthesis from *Centella asiatica* L plant extract are displayed in figure3. In the UV-VIS spectra the appearance of one or more peaks in the region from 200 to 1100 nm is a clear indication of the presence of unsaturated groups and heteroatoms such as S, N, O . The spectrum for Pt-NPs synthesis shows four peaks. This confirms the presence of organic chromophores within the Pt-NPs from *Centella asiatica* L. The applications of UV-visible spectrophotometry in the analysis of complex media is restricted by the difficulties in assigning the absorption peaks to any particular constituents in the system. The formation of Pt NPs was confirmed by a continuous absorption spectrum in the range of 200-1100 nm in UV-visible spectrophotometer. It can be seen that there is no distinct Surface Plasmon Resonance (SPR) occurs in this case and a continuous absorption is seen, which is in accordance with the earlier reports¹⁰

These absorption bands formed are attributed for the presence of flavonoids and its derivatives. The flavonoids spectrum typically consist of two absorption maximum in the range of 230-285 nm (band I) and 300-350 nm (band II). The specific position and relative intensities of these maxima give valuable information on the nature of the flavonoids. This result obtained are compared with the previous literature on *Acorus calamus*¹¹

The X-ray structural diffraction pattern of the Pt-NPs produced using *Centella asiatica* L was proved and confirmed by the characteristic peaks observed in the XRD images for platinum (Figure 8). The results of XRD patterns were interpreted using PCPDF WIN software. The XRD pattern recorded for Pt-NPs showed intense peaks in the whole spectrum of 2θ values ranging from 20 to 80. Intense peaks were observed at 10.5118, 25.3164, 28.5518, 37.9012, 47.9582, 53.8868, 54.9410, 62.5909, 69.7670, 75.2214 corresponding to (H₂PtCl₆) (Table 4) whereas any peaks originating because of potential platinum oxide interference could not be observed and it could not be confirmed that the entire chloroplatinic acid was converted to nanoplatinum. The broadening of the Bragg peaks observed indicates the formation of nanoparticles. The average grain size of the Pt-NPs formed in the bio-reduction was determined using the Scherrer equation ($D = 0.9\lambda / \beta \cos \theta$) and estimated as 11 nm. The XRD pattern clearly explains the crystalline structure of the Pt-NPs formed from plant extract. It seems that active secondary metabolites present in the leaf extract could be responsible for the platinum ions reduction. Previous studies have reported Pt-NPs to exhibit a brownish-red color in water due to the excitation of surface Plasmon vibrations in metallic nanoparticles¹². Thus, change of color in our study is indicative of platinum ions reduction by plant extract.

SEM images of the surface morphology of Pt-NPs can be visualized in Figure 9. The SEM images showed the aggregates of reduced platinum nanoparticles. This aggregates formation may be due to the high temperature and

the components present in *Centella asiatica* L leaf extract. Based on SEM images, the Pt particle size seems large because of the occurrence of agglomeration. Meanwhile, an analysis by using X-ray diffraction patterns shows that the size of the Pt crystals is nanoscale for all precursor concentrations. This difference is due to the X-ray diffraction pattern analysis that describes the size of the Pt crystals contained in Pt particles, because Pt particles are formed from collections of Pt crystals¹³

The nanoparticles produced were much larger with the majority of nanoparticles being rectangular and some other shape. The nanoparticles were aggregated, and thus it was very difficult to distinguish one shape from the other which supports with the observation. It appeared as though there was some extrapolymeric substance that coated the nanoparticles kept them closely attached to each other, rectangular nanoparticles were produced by the bioreduction of H₂PtCl₆ and these appeared to be monodispersed and varying in size. The results indicated that in addition to pH and temperature, the oxidation state of the platinum salt played an important role in the mechanism and formation of the nanoparticles though the size and shape of the particles was uncontrolled.

CONCLUSION

From the results, it was confirmed that Pt-NPs synthesized from *Centella asiatica* L leaf extract are responsible for the biological activities that is useful for natural health. Further studies on Pt-NPs synthesized from *Centella asiatica* L leaf extract needed to take the research forward for further exploration.

CONFLICTS OF INTEREST:

The author have declared no conflicts of interest

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