



## Green-synthesis of Platinum Nanoparticles using Olive Leaves Extracts and its Effect on Aspartate Aminotransferase Activity

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### Abstract

This study aims to synthesize Platinum nanoparticles (Pt NPs) using Olive leaves extract's environmentally friendly approach. UV-Visible spectroscopy has been used to follow the turning of Pt +4 ions to Pt<sup>0</sup> NPs. X-ray diffraction (XRD) has been used to investigate the as-synthesized Pt NPs and its cubic face-centered structure crystallinity. The average size of nanoparticles was 9.2 nm, calculated by transmission electron microscopy (TEM). The activity of as-synthesized Pt NPs has been examined by inhibition of Serum Aspartate Aminotransferase (AST) level in patients with chronic liver disease and control group. The AST (mean= 12.8051.642) demonstrate a very significant rise ( $p < 0.01$ ) in the results. In chronic liver patients with Pt NPs, serum AST activity was significantly lower ( $p < 0.01$ ) than in patients without Pt NPs (mean=13.4582.360). The present study concluded that Platinum nanoparticles play a great role in inhibition of AST.

*Keywords: Platinum nanoparticles, Olive leaves extract, TEM, AST.*

### 1. Introduction

Nanomaterials are among the most active research areas in recent materials science [1-4]. In the current, nanomaterials have a significant consideration owing to their applications in the biomedical, physicochemical fields, drug delivery, sensing, imaging, and chemotherapy [5-9]. The significant role of noble metal nanoparticles in physics, chemicals, materials science, biological and medicinal fields has been widely estimated [10-14]. Platinum has high density silver-white precious metal, a high surface, a high melting point (1769 °C), and high resistance to corrosion and chemical attacks. It is an essential catalyst for automotive emission reduction, hydrogen storage, membrane exchange cells for protons, and direct methanol cells. The recent behavior of platinum nanoparticles is close to NADH oxidization, and coenzyme Q has been

reduced [15]. Besides, for the preparation of organic dyes, nanoparticles platinum is used [16]. Different methods such as chemical precipitation, the sol-gel method, pyrolysis, soil phase, hydrothermal synthesis, vapour, and electro-deposition have been developed to synthesize Pt NPs. The techniques mentioned above have some limitations like high energy requirement, multi-step process, and unsafe chemicals. The green synthesis technique was used to overcome these problems where. It is characterized as is simple, low cost, eco-friendly. This eco-friendly approach helps synthesize nanoparticles of various types, sizes, and applications, such as antimicrobial, anticancer, antioxidant, larvicide, and antibiofilm. [17–22]. In this work, we report synthesis of Pt-NPs using Olive leaves extract and study the inhibitory effects of these green synthesis nanoparticles on the AST in sera of patients with chronic liver disease.

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## 2. Experimental Part

Olive leaves, Deionized water, and hydrated hexachloroplastic acid [H<sub>2</sub>PtCl<sub>6</sub>.6 (H<sub>2</sub>O)]. All chemical compounds have been reagent analytic test scores and have been used without further cleaning from Sigma-Aldrich.

### 2.1. Olive leaves extract preparation

Olive leaves have been collected and cleared from dust particles several times with de-ionized water. The leaves were turned into powder after drying in an oven at 80 °C for 3 hours, then crushed in a mortar. Boiling 25 g of the leaves powder in 200 mL of de-ionized water at 95 °C for 25 min. The leaves extract solution was made. The extract was refrigerated, taken, and processed in 4 OC. The solution was used within a week of planning [23].

### 2.2. Green Synthesis of platinum nanoparticles

The chemical source of Platinum NPs used in this experiment was the dried hydrated hexachloroplastic acid [H<sub>2</sub>PtCl<sub>6</sub>.6 (H<sub>2</sub>O)]. 15 ml of Olive leaves extract was added slowly (drop by drop) to 45 ml, 1 mM of [H<sub>2</sub>PtCl<sub>6</sub>.6 (H<sub>2</sub>O)] for 60 min under ultrasonic sonication, the PH was 5.5 at 15 °C. Platinum nanoparticles were first characterized by a change in color visually at a different time of incubation (10, 20, and 30 minutes) in the reaction mixture containing [H<sub>2</sub>PtCl<sub>6</sub>.6 (H<sub>2</sub>O)] and Olive leaves extract. A brown solution has been observed after 30 minutes. This ensures that platinum nanoparticles are formed in the solution [24].

### 2.3. Specimens Collection

A total sample of 25 patients with liver disease was acquired from the Wasit region/Iraq, which corresponds to 25 controls included in this study. These samples were taken from all the Patients and control by plastic disposable syringes and were allowed clotting at room temperature for 15 minutes from the patients and controls. The ingredients were separated at 700 g for 10 minutes by centrifugation. The serum samples have isolated and placed immediately in plastic tubes and kept at (-20 °C) to later study.

### 2.4. In vitro of Pt-NPs on AST

In this study, Pt nanoparticles (2 µg/0.5 ml) was added to 1 ml of serum from each sample (1:2). They were mixed for 300 sec, and the amount of AST was measured.

### 2.5. Statistical analysis

An analysis of the data was performed through Microsoft Office (SPSS version 24), and the results are analyzed using One-way ANOVA. Result values were expressed as average ± standard deviation (SD), the values  $p < 0.05$  were regarded statistically significant.

## 3. Results and discussions

The X-ray diffraction (XRD) patterns of platinum nanoparticles synthesized by extracts of olive leaves shows in Figure 1. The powdered platinum-nanoparticle XRD spectrum has three different high peak levels of diffraction at 39.3°, 45.8°, and 67.33° refers for index planes (111), (200) and (220) respectively. The values corresponded to the cubic face-centered structure. With the achieved XRD pattern, the crystalline structure of the formed Pt NPs was verified. Debye-Scherrer formula [25-29] can estimate crystalline sizes of Pt NPs and is found equal to 8.3 nm. The broadening of the peak in XRD is a clear indication of Particle formation at the nanoscale.

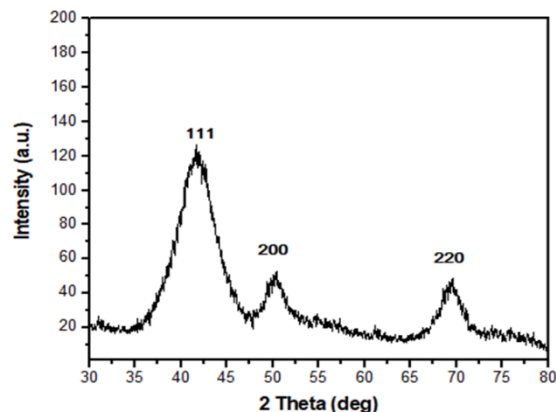


Fig 1. XRD pattern of Pt NPs eco-friendly synthesized

TEM was selected for the characterization of Pt NPs because it provides more excellent resolution and more accurate particle-size than other technologies such as scanning electron microscopy (SEM). PtNPs with high-resolution TEM synthesized green shows in Figure 2 (a), and figure 2 (b) shows the randomly determined particle distribution of the TEM image. After characterizing the TEM samples, the actual size, type, and morphology of nanostructures are being confirmed. Furthermore, the photos show that while some Pt NPs are mostly spherical, those who influence with strong cubic form limits. These studies showed the average nanoparticles size and distribution. The most important characteristic of the distribution of platinum nanoparticles is the homogeneous distribution that appeared in the

measurement, and the reason for this is due to the accuracy of the olive leaf extract, which works to reduce the surface tension (surfactant) which prevents agglomerations and this is confirmed by the TEM measurement. The TEM picture shows that the prepared Pt NPs are very scattered, spherical, compact, and even in the average size of 9.2 nm.

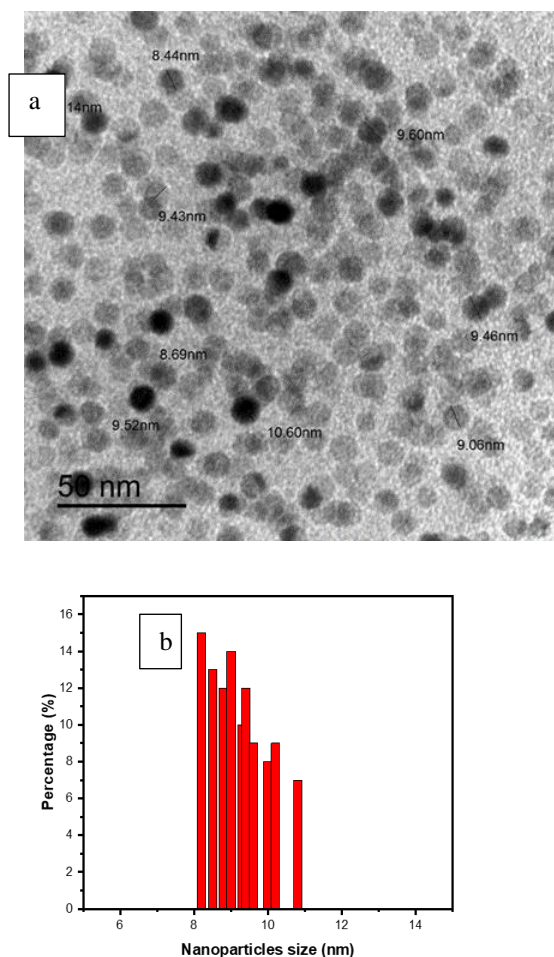


Fig.2. a) TEM image of Pt NPs eco-friendly synthesized. b) Distribution of Pt NPs by TEM

After the Pt<sup>4+</sup> to Pt<sup>0</sup> nanoparticles reduction process, the UV – Vis spectrometer was used. Color changes were monitored, and the solution change from yellow to yellowish-brown, suggesting that H<sub>2</sub>PtCl<sub>6</sub> was reduced, and platinum nanoparticles were produced. Reflected the record arousal of surface plasma vibration in the platinum particles show in Figure 3. The value of surface Plasmon Resonance (SPR) in the olive extract leaves for platinum nanoparticles at 358 nm depends on its shape and scale [30].

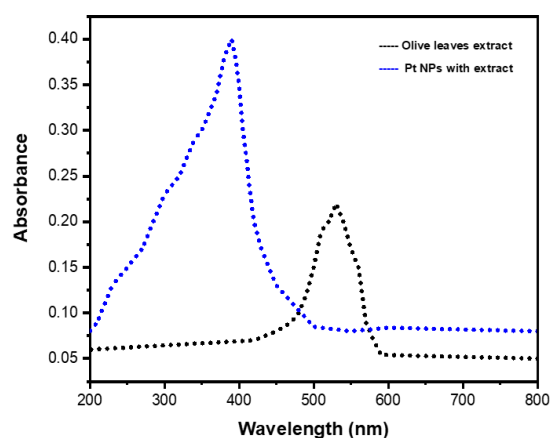
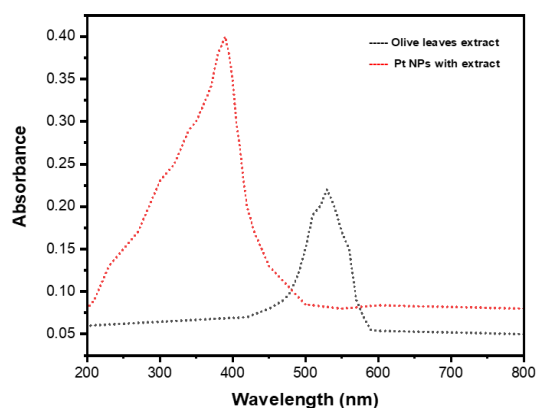


Fig 3. UV – Vis of produced platinum nanoparticles

R.Karthik et.al reported the electrochemical detection of hydrazine, Pt NPs were synthesized using *Quercus glauca* (Qg) leaves extract in result Pt NPs have spherical shape and 5.15nm size [15]. John Leo et.al Using aqueous extracts from the water hyacinth plant as efficient reducing and stabilizing agents, platinum nanoparticles (Pt-NPs) were synthesized and Pt-NPs spherical in shape with size 3.74 nm [31]. V. SriRamkumar et.al reported how PtNPs were synthesized by using an aqueous extract of the Indian brown seaweed *Padina gymnospora*, and their catalytic activity was tested with a polymer Polyvinylpyrrolidone (PVP) as a PVP/PtNPs nanocomposite for antibacterial, haemolytic, cytotoxic (*Artemia salina*), and antioxidant characteristics. In result was found PtNPs octahedral in shape and 5-50 nm size [32]. In our study, the PtNPs shown cubic facially in shape with small average size 9.2 nm and homogeneous distribution due to Olive leaves extracts.

Groups	Aspartate transaminase Mean $\pm$ SD	p-value
Control	12.805 $\pm$ 1.642	p<0.01
Patients without Pt NPs	39.114 $\pm$ 2.442	p<0.01
Patients with Pt NPs	13.458 $\pm$ 2.360	p<0.01

Table 1: Aspartate aminotransferase level in sera of controls and chronic liver patients

### 3.1. Effect of Pt NPs on AST enzyme activity

The effect of Pt nanoparticle on AST activity was inspected in the serum levels of patients with chronic liver disease show in Table 1 and figure 4. The results showed the inhibition of the AST activates by Pt-nanoparticle. In amino acid metabolization and urea tricarboxylic acid cycles, AST plays an important role. [22]. Serum AST (mean=39.114 $\pm$ 2.442) in chronic liver disease patients without Pt NPs, the results show a highly significant increase (p<0.01) in the serum levels of the AST compared with the control group (mean = 12.805 $\pm$ 1.642). The results also have shown a highly significant decrease (p<0.01) in the serum levels of AST activity in chronic liver patients with Pt nanoparticle (mean=13.458 $\pm$ 2.360) compared to serum patients without Pt nanoparticle. We proposed that nanoparticles molecule changes the active parts of amino acids on the AST, owing to lessening

### 4. Conclusions

In conclusion, An Eco-friendly Method (green synthesis) was used to Synthesis zero-valent Platinum Nanoparticles with the extract of olive leaves is an alternative environmentally. The platinum ion reduction agents include acid ascorbic, gallic acids, terpenoids, various proteins, and amino acids found in the olive leaf extract. The average Pt-NPs was 9.2 nm with a cubic facially centered structure. The effect of Pt NPs on AST behavior has been investigated. The results showed the high impact of the Pt NPs on the enzyme activity. Finally, the prepared Pt NPs works effectively against enzymes.

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affinity of active sides of an enzyme or the change in the p<0.01= highly significant.

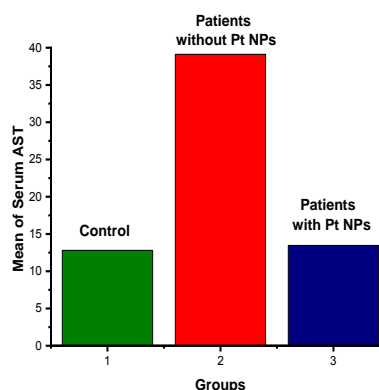


Fig 4. AST level in samples sera of control and liver patients.

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