Journal of Photochemistry & Photobiology, B: Biology 166 (2017) 246–251

Contents lists available at ScienceDirect

Journal of Photochemistry & Photobiology, B: Biology

journal homepage: www.elsevier.com/locate/jphotobiol

Facile and green synthesis of phytochemicals capped platinum nanoparticles and in vitro their superior antibacterial activity

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ARTICLE INFO

Article history:
Received 28 October 2016
Received in revised form 5 December 2016
Accepted 10 December 2016
Available online 21 December 2016

Keywords:
Taraxacum laevigatum
Green synthesis
Platinum nanoparticles
Antibacterial activity

ABSTRACT

The increase in the severe infectious diseases and resistance of the majority of the bacterial pathogens to the available drug is a serious problem now a day. In order to overcome this problem it is necessary to develop new therapeutic agents which are non-toxic and more effective to inhibit these microbial pathogens. For this purpose the plant extract of highly active medicinal plant, *Taraxacum laevigatum* was used for the synthesis of platinum nanoparticles (PtNPs) to enhance its bio-activities. The surface plasmon resonance peak appeared at 283 nm clearly represent the formation of PtNPs. The results illustrate that the bio-synthesized PtNPs were uniformly dispersed, small sized (2–7 nm) and spherical in shape. The green synthesized PtNPs were characterized by UV–vis spectroscopy, XRD, TEM, SEM, EDX, DLS and FTIR. These nanoparticles were tested against gram positive bacteria (*Bacillus subtilis*) and gram negative bacteria (*Pseudomonas aeruginosa*). The bio-synthesized PtNPs were examined to be more effective against both of the bacteria. The results showed, that the zone of inhibition of PtNPs against *P. aeruginosa* was 15 (±0.5) mm and *B. subtilis* was 18 (±0.8) mm. The most significant outcome of this examination is that PtNPs exhibited strong antibacterial activity against *P. aeruginosa* and *B. subtilis* which have strong defensive system against several antibiotics.

1. Introduction

Noble metal nanoparticles have potent applications in numerous fields i.e. chemical, medical, electronics and biological [1–3]. The effective applications of nanoparticles greatly depend upon their size, shape, morphology and dispersions [4–7]. Platinum is a precious noble metal and its nanoparticles have strong applications in various fields. The metallic platinum compound i.e. *cis*-diammine dichloro platinum has been applied as a cancer drug [8]. PtNPs have been used in the fuel cells and hydrogen storage materials [9,10]. They were found to have significant catalytic applications than bulk materials [11]. The PtNPs were also proved as the most useful catalyst in the proton membrane exchange fuel cells [12]. They are also widely used in most of the hydrogenation reactions i.e. hydrogenation of *o*-chloronitrobenzene and cinnamaldehyde [13,14]. In addition, PtNPs being applied for the synthesis of organic dyes [15]. Moreover, the several complexes of platinum have been used against different gram positive and gram negative bacterial species.

A series of conventional protocols have been used in preparation of noble metal nanoparticles i.e. UV irradiation reduction [16], laser ablation [17], electrolysis method [18], thermal decomposition [19], microwave processing [20], ion implantation [21] and chemical reduction [22,23] etc. All these methods have some major drawbacks i.e. use of expensive and hazardous chemicals etc. which has increase the attention of the researchers to introduce environment friendly alternative procedures using biological system in preparation of nanoparticles. It is well known that biological systems have a strong efficiency for production of spherical shape, small size and highly stable nanoparticles. Keep it in mind, the researchers introduce medicinal plants as an alternate for the preparation of nanoparticles instead of hazardous chemicals because it is non-toxic, cheap and easily available [24–29]. The phytochemicals of the plants play an important role in the synthesis of nanoparticles. The water soluble organic moiety of the medicinal plants is not only use for the reduction of the nanoparticles but also stabilize the prepared nanoparticles. The current reports demonstrated that plants extract are more advantageous for the preparation of metal nanoparticles over other conventional methods because they consists of high concentrations of biomolecules i.e. terpenoids, phenols, alkaloids, flavonoids, quinines, tannins etc. which are responsible for the reduction and stabilization of metal nanoparticles.

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http://dx.doi.org/10.1016/j.jphotobiol.2016.12.016
1011-1344/

Noble metal nanoparticles (NMNPs) are promise antibacterial agents possessing strong antibacterial efficiency with negligible bacterial resistance against them [30]. It has been previously reported that the ions of noble metals damage the bacterial DNA, cell membrane, critical enzymes and destroy bacteria by a process called respiratory burst mechanism [31,32]. Moreover, the nanoparticles of noble metals have the ability of producing reactive oxygen species which are responsible for inhibition of pathogenic microbes.

In the present work, highly dispersed, spherical shape, small size and uniformly distributed platinum nanoparticles were synthesized using the plant extract of *Taraxacum laevigatum* as reducing and capping source. The plant of *Taraxacum laevigatum* has phenolic biomolecules which are the main agents for the reduction and stabilization of PtNPs. The PtNPs prepared at optimized conditions i.e. 10 mL extract concentration and 90 °C temperature were spherical in shape, small sized and uniformly distributed. The prepared PtNPs were screened for their bio-medical application i.e. antibacterial activity. The result demonstrated that PtNPs have strong efficiency towards both gram positive and gram negative bacterial inhibition.

2. Materials and Method

2.1. Preparation of Plant Extract

The *Taraxacum laevigatum* plant materials were collected from Pakistan and washed with distilled water to remove dust particles and other impurities and then shade dried. 5 g of the plant powdered was extracted with 100 mL Milli-Q water. The suspended plant materials in deionized water were heated at 70 °C for 40 min with continuous stirring (500 rpm). Finally, the water extract was filtered through Whatman filter paper (Whatman filter paper No. 3). The clear supernatant obtained was stored at 4 °C for further use.

2.2. Synthesis of PtNPs Using Taraxacum laevigatum Plant Extract

For the synthesis of PtNPs, 10 mL of *Taraxacum laevigatum* plant extract was added to 50 mL of 0.01 M aqueous solution of H_2PtCl_2 in 100 mL beaker. The change in color took place from yellow to brown within 10 min when heated at 90 °C. After the preparation of PtNPs it was centrifuged and then dried in 6ES freeze drier.

2.3. Antibacterial Activity of PtNPs

The PtNPs were tested against one gram positive and one gram negative bacteria. The two bacterial species i.e. *Bacillus subtilis* (*B. subtilis*) and *Pseudomonas aeruginosa* (*P. aeruginosa*) were used in this study. Strains were obtained from School of Science Beijing University of Chemical Technology, China 100029, Lab. No. 1104.

The antibacterial effectiveness of PtNPs was observed by previously reported protocol [33]. The two bacterial strains were grown in agar broth at 37 °C for 24 h in an incubator. Subsequently, the inocula of the particular bacterium were splashed on the Muller Hinton agar plates. Wells of 6 mm in diameter were made with the help of sterile cork borer on the agar plates. Alternatively 1 mg PtNPs were dissolved in 1 mL doubly distilled water and the wells were filled with 50 μL of PtNPs. Moreover, the plates were incubated for 24 h at 37 °C and the zones of inhibition were carefully measured. In this assay streptomycin was used as a standard.

2.3.1. Minimum Inhibitory Concentration (MIC)

The MIC of PtNPs was performed by Serial dilution protocol. PtNPs of different concentrations (25 μg to 120 μg) were taken and mixed with 1 mL of two bacterial solutions in uncontaminated test tubes. After that the test tubes were placed in a shaking incubator for 24 h at 37 °C. The test tube containing bacteria and growth media without PtNPs was used as a negative control.

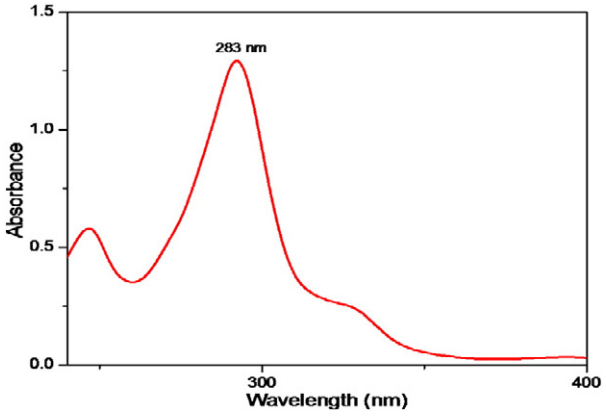

Fig. 1. UV–vis spectra of bio-synthesized PtNPs using 10 mL *Taraxacum laevigatum* plant extract at 90 °C temperature.

2.4. Characterization

The biologically synthesized PtNPs were examined initially by UV–vis spectrophotometer (Shimadzu UV-2400). The X-ray diffraction analysis of PtNPs was done on a (Rigaku D/Max 2500 VBZ+/PC) diffractometer. HRTEM of PtNPs was analyzed on a JEM-3010 microscope. FT-IR spectrum was analyzed at ABB MB3000 spectrophotometer. The element distribution maps and EDX were examined by Hitachi S-4700 SEM. Dynamic light scattering technique (DLS) was applied to investigate the size distribution of platinum nanoparticles and their stability by zeta potential values (HORIBA Zeta sizer SZ100).

3. Results and Discussions

3.1. UV–Visible Spectral Analysis

UV–vis spectroscopy is an absorption spectroscopy which is widely used to confirm the synthesis of nanoparticles and nanocomposites. Every element have their own specific excitation energy and the UV–vis radiations are highly energetic which are enough for the excitation of valence electron of any element to the high energy state, so as a result the peak appeared for that element in the UV–visible region.

The synthesis of PtNPs was first confirmed by UV–visible spectroscopic analysis as shown in Fig. 1. Due to the presence of free electrons, every metal gives rise to surface plasmon resonance (LSPR) peak [34]. It is previously reported that the intensity and specific position of LSPR peak is dependent on the size and shape of the nanoparticles formed [6]. The peak originated at 283 nm clearly illustrates the formation of PtNPs.

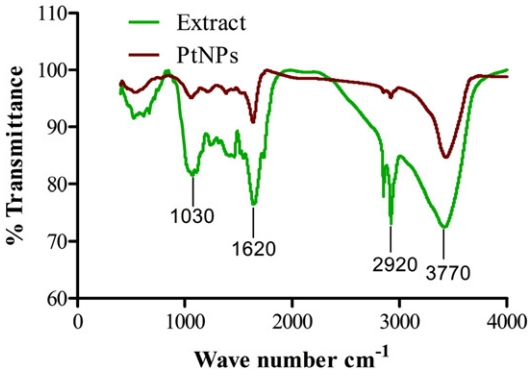

Fig. 2. FT-IR spectra of plant extract and plant mediated PtNPs.