Contents lists available at ScienceDirect



Journal of Photochemistry & Photobiology, B: Biology

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



Biosynthesis of polyphenol-stabilised nanoparticles and assessment of anti-diabetic activity



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ARTICLE INFO

Article history: Received 27 November 2016 Received in revised form 17 February 2017 Accepted 21 February 2017 Available online 24 February 2017

Keywords: W. somnifera Pt NPs Controlled synthesis Anti-diabetic activity

1. Introduction

Nanotechnology focuses primarily on the synthesis of nanoparticles (NPs) using biological, physical and chemical methods. Synthesis of NPs by chemical and physical methods produces environmental toxins. Researchers have focused on synthesising NPs via biological methods, including green synthesis and microbe-mediated synthesis, to avoid pollution [1]. Green synthesis uses plants and plant parts, and microbe-mediated synthesis uses bacteria, fungi and yeast [2].

This study focused on plant-mediated synthesis of NPs using *Whitania somnifera* leaves, because of their previously reported health benefits [3] *W. somnifera* has been utilised to treat coronary artery disease, heart failure, angina pain, etc. [4]. *W. somnifera* has several common names, such as Ashwagandha [5], Indian ginseng [6] and poison gooseberry [7]. Metal NPs have been used in biomedical applications as sensors, catalysts and chemical sensors and in pharmaceutical applications [8,9]. Platinum (Pt) is also used in the synthesis of NPs and in the treatment of several medical disorders, such as cancer and diabetes [10].

Diabetes has becomes a worldwide threat, even in south Asian countries [11]. Streptozotocin (STZ) is an antibiotic derived from the bacteria *Streptomyces achromogenes* that has been used to induce diabetes experimentally [12]. STZ is incorporated by hepatocytes and pancreatic

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ABSTRACT

Green synthesis of nanoparticles (NPs) is a major developing field. In this study, we utilised *Whitania somnifera* leaves as a green source to synthesise platinum (Pt) NPs. Synthesised Pt NPs resulted from controlled synthesis of 12 nm spherical particles. The synthesised Pt NPs were subjected to anti-diabetic applications resulting in a significant decrease in plasma glucose levels after injecting the Pt NPs into streptozotocin-induced diabetic rats. © 2017 Elsevier B.V. All rights reserved.

> beta cells expressing glucose transporter 2. Hepatocytes can protect themselves from STZ toxicity, but beta cells are selectively damaged and die [13]. A few medications such as pregabalin and gabapentin are available to treat diabetic neuropathy, but they present various side effects [14]. To overcome these side effects, researchers have begun to focus on biological components, such as quercetin, curcumin and naringin, which are easily isolated from various plant sources [15,16]. Very few reports are available on the green synthesis of Pt NPs and their potential anti-diabetic activity. In this study, we used an ecofriendly approach to synthesise Pt NPs and evaluated their effects in STZ-induced rats. Our objective was to synthesise Pt NPs using a *W. somnifera* leaf extract and use them to treat diabetic rats.

2. Materials and Methods

2.1. Sources

Chloroplatinic acid and STZ were procured from Sigma-Aldrich (Beijing, China). Double distilled water was utilised in all experiments.

2.2. Plant Collection

W. somnifera powder was collected from local markets. Adult male Sprague–Dawley rats (weight, 150–200 g) (Slac Laboratory Animal Co. Ltd., Shanghai, China) were housed at 45–55% relative humidity and 24 °C under a 12 h dark and 12 h light cycle. The animals were fed pelleted chow. All anti-diabetic activity testing was performed during



Fig. 1. Ultraviolet-visible spectroscopy analysis of platinum nanoparticles (Pt NPs) synthesised via Whitania somnifera aqueous leaf extract.

the daytime. The study protocol and the animal use for this experiment were approved by the Ethical Committee.

2.3. W. somnifera Leaf Extract

W. somnifera leaves were collected, sun dried, ground to a powder and stored at 4 °C until use. Approximately 30 g powdered material were immersed in 100 mL distilled water, heated at 60 °C for 3 h and monitored using UV–visible spectroscopy. Once the Pt NPs were confirmed by their UV–visible absorbance range, the reaction was stopped and centrifuged at 3000 rpm for 30 min. This process was repeated three times to remove secondary metabolites from the plant source. After centrifugation, the supernatant was discarded, and the pellet was powdered in a furnace at 450 °C for 4 h.

2.4. Characterisation of Pt NPs

The synthesised Pt NPs were subjected to various analytical techniques, such as UV–visible spectroscopy, X-ray diffraction (XRD) for crystalline studies and Fourier transform infrared spectroscopy (FT-IR) to identify the functional groups. Transmission electron microscopy/energy dispersive X-ray (TEM/EDAX) was used for the morphological and elemental analyses. A Horiba NP analyser was used to determine the stability of the zeta potential.

2.5. Anti-diabetic Studies

Diabetes was induced in male Sprague–Dawley rats by a single intravenous injection of STZ (60 mg/kg). Animals with fasting plasma glucose levels >150 mg/dL (diabetic) after 48 h were selected for the study. We followed a previous method [17] with slight modifications as follows; we used the following four groups consisting of six rats each:

Group 1: Normal control rats; received only 10 mL/kg distilled water **Group 2:** STZ-induced rats

Group 3: STZ-induced rats administered 10 mL/kg W. somnifera extract

Group 4: STZ-induced rats administered 1 mg/kg Pt NPs

Blood glucose levels were examined in all groups at 1, 7, 14, 21 and 28 days. The animal experiment was approved by Animal Welfare Committee of Fujian Medical University (Fuzhou, China).



Fig. 2. X-ray diffraction studies of green-synthesised platinum nanoparticles (Pt NPs) and the crystalline data.

2.6. Statistical Analysis

The data are expressed as means \pm SD using GraphPad Prism software ver. 5.0 (GraphPad Inc. La Jolla, CA, USA). A *P*-value < 0.05 was considered significant.

3. Results and Discussion

3.1. Pt NP Characterisation

W. somnifera-mediated synthesis of Pt NPs was monitored using UV–visible spectroscopy. The highest absorbance peak was observed at 267 nm when the colour changed from pale brown to dark brown, indicating that Pt^{4+} was converted into Pt^0 [11] (Fig. 1).

The XRD analysis of Pt NPs revealed a crystalline structure with $2\Theta = 38, 43, 63$ and 82, which indexed as 111, 200, 220 and 311, respectively, resulting in a face-centred cubic structure and the formation of Pt NPs (Fig. 2). The size of the crystals was determined to be 14 nm using the Scherrer formula:

 $D = K\lambda/\beta \cos\theta$,

where D is the particle size, K is Scherer's (0.94) constant, $2dsin\theta = n\lambda$ is Bragg's equation, λ is the wavelength, β is the full width at half maximum value, and θ is the diffraction angle.



Fig. 3. Fourier transform infrared analysis of platinum nanoparticles (Pt NPs) synthesised via Whitania somnifera aqueous leaf extract.

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