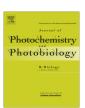
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Mycosynthesis: Antibacterial, antioxidant and antiproliferative activities of silver nanoparticles synthesized from *Inonotus obliquus* (Chaga mushroom) extract



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ABSTRACT

In the present study, silver nanoparticles (AgNPs) were rapidly synthesized from silver nitrate solution at room temperature using *Inonotus obliquus* extract. The mycogenic synthesized AgNPs were characterized by UV–Visible absorption spectroscopy, Fourier transform infrared (FTIR), scanning electron microscopy (SEM) with energy dispersive spectroscopy (EDS), transmission electron microscopy (TEM) and atomic force microscopy (AFM). SEM revealed mostly spherical nanoparticles ranging from 14.7 to 35.2 nm in size. All AgNPs concentrations showed good ABT radical scavenging activity. Further, AgNPs showed effective antibacterial activity against both gram negative and gram positive bacteria and antiproliferative activity toward A549 human lung cancer (CCL-185) and MCF-7 human breast cancer (HTB-22) cell lines. The samples demonstrated considerably high antibacterial, and antiproliferative activities against bacterial strains and cell lines.

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1. Introduction

The "green synthesis" of metal nanoparticles has received great attention due to their unusual optical, chemical, photochemical, and electronic properties [1]. Metal nanoparticles can be synthesized via chemical [2,3] and biological methods. Chemical methods for metal nanoparticle fabrication usually involve toxic materials that are expensive and potentially harmful to the environment [4]. However, green synthesis of nanoparticles is an easy, inexpensive, efficient and eco-friendly biological method of biosynthesis of AgNPs. Such methods use plants [5–7], bacteria [8,9], fungi [10,11] or yeast [12,13], which are known to reduce silver ions into silver nanoparticles both extra and intracellularly [14–16], as well as mushrooms such as *Volvariella volvacea* [17], *Pleurotus sajor* [18], *Pleurotus florida* [19], *Ganoderma lucidum* [20], *Agaricus bisporus* [21]. In this study, we used extract of the *Inonotus obliquus* to synthesis of AgNPs.

Mushrooms are known to have anti-inflammatory, antitumor, antiviral, antivaterial, hepatoprotective and hypotensive activities in biological systems [22–24]. Many varieties of naturally occurring mushrooms have long been known to have promising

antioxidant and anticancer properties and prolong longevity [25], as well as to contain antitumor compounds [26,27]. Accordingly, a variety of edible mushrooms have been taken as vitamin and mineral supplements.

I. obliquus (family: Hymenochaetaceae) is a black mushroom that grows on birch trees in northern climates such as Russia [28]. I. obliquus mushrooms have been used as a folk remedy in Russia and Eastern Europe since the 16th century for a wide variety of human diseases without any intolerable toxic side effects [29,30]. These mushrooms are also used as traditional medicine for the treatment of diabetes [31,32]. Additionally recent studies, have reported that the polyphenolic compounds produced by I. obliquus can protect cells against oxidative stress [32]. These mushrooms have been reported to show therapeutic benefits including anti-inflammatory, immune-modulatory and hepatoprotective effects [33]. Despite its popular use, the mechanism underlying the pharmacological activity of I. obliquus is yet to be elucidated.

2. Material and methods

I. obliquus mushrooms were obtained from a forest area of Siberia, Russia. Silver nitrate (AgNO₃) was procured from Sigma–Sldrich Chemical Pvt. Ltd., Seoul, South Korea.

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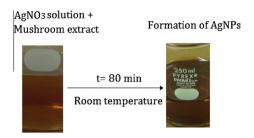


Fig. 1. Color change of *Inonotus obliquus* extract containing silver before and after synthesis of AgNPs. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

2.1. Preparation of mushroom extract

A total of 10 g mushrooms were washed repeatedly with distilled water to remove any organic impurities. The cleaned mushrooms were then crushed into small pieces with a sterilized knife and placed into a 500 mL beaker containing 200 mL double distilled water and thoroughly stirred for about half an hour. This solution was then filtered through whatman filter paper no. 41 and stored $4\,^{\circ}\text{C}$.

2.2. Mycosynthesis of AgNPs

An aqueous solution of silver nitrate (1 mM) was prepared and used for the synthesis of AgNPs. Specifically, 5 mL of mushroom extract was added into 95 mL of 1 mM silver nitrate to reduce Ag⁺ to Ag⁰. This solution exposed to room temperature, which resulted in a change in color from orange color to dark orange within 80 min.

2.3. Characterization

UV–Vis spectral analysis was performed using a Cary-4000 spectrophotometer. The morphology of the prepared AgNPs was observed by SEM (Hitachi, S-4800), EDX (Horiba, 6853-H), TEM

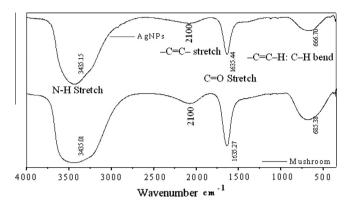


Fig. 3. FTIR spectra of *Inonotus obliquus* extract and AgNPs from *Inonotus obliquus* extract

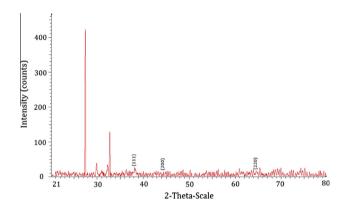


Fig. 4. XRD pattern of green synthesized AgNPs from Inonotus obliquus extract.

(Hitachi, H-7100) and AFM (Veeco dimension 3100 SPM). The structure and composition of AgNPs were analyzed by XRD (AXS D8 Advance). Further characterization was accomplished using FTIR (Bruker Germany).

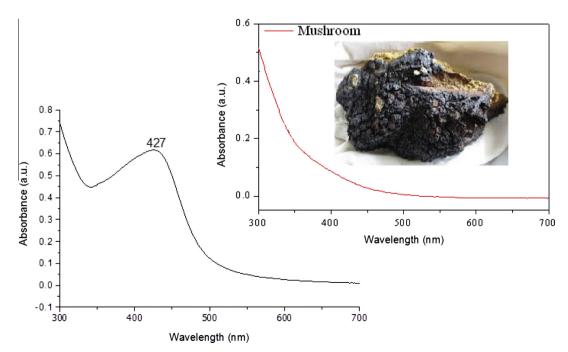


Fig. 2. UV-Visible spectrum of AgNPs synthesized by treating 1 mM aqueous AgNO3 solution with Inonotus obliquus extract after 80 min.

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