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SYNTHESIS OF SILVER NANOPARTICLES FROM FUNGI ISOLATED FROM MARINE ECOSYSTEM

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Abstract

The biosynthesis of aqueous Ag⁺ ions by the fungus extract has been demonstrated. The findings of the present study has been reported as green chemistry approach using a fungi in the synthesis of silver nanoparticles at room temperature without using any harmful reducing agents such as sodium borohydrite or hydroxylamine hydrochloride and any capping or dispersing agent. From this investigation, we have concluded that the fungal biodiversity in mangrove ecosystem, *Aspergillus* and *Penicillium* was the common fungal genera among the isolated from the study period. Fungi play an important role in decomposition of natural substrates in mangrove ecosystem. The fungi isolated from mangroves are mainly used in enzyme technology, biochemical, agricultural, pharmaceutical, molecular biology and other applied research fields.

Keywords: Marine fungi, Silver Nanoparticles , *Aspergillus* and *Penicillium*

Introduction

The need for biosynthesis of nanoparticles rose as the physical and chemical processes were costly. So in the search of for cheaper pathways for nanoparticle synthesis, scientists used microorganisms and then plant extracts for synthesis. Nature has devised various processes for the synthesis of nano- and micro- length scaled inorganic materials which have contributed to the development of relatively new and largely unexplored area of research based on the biosynthesis of nanomaterials (Mohanpuria *et al.*, 2007). Biosynthesis of nanoparticles is a kind of bottom up approach where the main reaction occurring is reduction/oxidation. The microbial enzymes or the plant phytochemicals with anti oxidant or reducing properties are usually responsible for reduction of metal compounds into their respective nanoparticles. The three main steps in the preparation of nanoparticles that should be evaluated from a green chemistry perspective are the choice of the solvent medium used for the synthesis, the Biomimetic Synthesis of Nanoparticles: Science, Technology & Applicability 5 choice of an environmentally benign reducing agent and the choice of a non toxic material for

the stabilization of the nanoparticles. Most of the synthetic methods reported to date rely heavily on organic solvents. This is mainly due to the hydrophobicity of the capping agents used (Raveendran *et al.*, 2002). Synthesis using bio-organisms is compatible with the green chemistry principles: the bio-organism is (i) eco-friendly as are (ii) the reducing agent employed and (iii) the capping agent in the reaction (Li *et al.*, 2007). Often chemical synthesis methods lead to the presence of some toxic chemical species adsorbed on the surface that may have adverse effects in medical applications (Parashar *et al.*, 2009). This is not an issue when it comes to biosynthesized nanoparticles as they are eco friendly and biocompatible for pharmaceutical applications.

Use of organisms to synthesize nanoparticles

Biomimetics refers to applying biological principles for materials formation. One of the primary processes in biomimetics involves bioreduction. Initially bacteria were used to synthesize nanoparticles and this was later

succeeded with the use of fungi, actinomycetes and more recently plants. Bio-reductant from bacteria, fungi, or plant parts + Metal ions (Maybe enzyme/ phytochemical) Metal nanoparticles in solution.

Materials and Methods

Production of silver nanoparticles

20gm of fungal mycelium was obtained from the liquid media used for the synthesis of silver nanoparticles. 20 ml of the cell free filtrate was brought and transferred to 10⁻³ M concentration silver nitrate Erlen Meyer flask and agitated at 25°C in darker conditions under normal pH. Simultaneously, control without silver ions was also run along with the experimental flasks.

UV visible studies

The reduction of silver ions was monitored by measuring the UV-VIS spectrum of the reaction medium at 24 hrs time interval by drawing 1cm of the samples and their absorbance was recorded at a resolution of 0.5m at 350-800nm using UV-VIS spectrophotometer – UV 2450(Shimadzu).

SEM Analysis

Thin films of the sample was prepared on a carbon coated copper grid by just dropping a very small amount of the sample on the grid, extra solution was removed using a blotting paper and then the film on the SEM grid was allowed to dry by putting it under a mercury lamp for 5 mins for emitting characteristic X-rays. These characteristic X-rays are used to identify

the composition and measure the abundance of elements in the sample.

Results and Discussion

Nanosilver formation:

The colour change occurred in the cell free extract when challenged with 1mM AgNO₃ changed colour from pale yellow to dark brown colour in 48 hrs and attained maximum intensity after 72 hrs with intensity increasing during the period of incubation indicative of the formation of silver nanoparticle. Control without silver ions showed no change in colour of the cell filtrates when incubated under same conditions.

UV – VIS spectral studies

Typical UV- VIS spectra of the reaction solution recorded at an interval between 0 – 72 hrs. Under normal pH 6.0 the change in light absorption profile of the medium and change in intensity of the brown colour during long term incubation (72hrs). In the filtrate obtained from the fungal extract, a new path was observed in the visible region of (440nm) and suggested that the organism reduced the silver nitrate to silver oxide as the growth of the organism's preceded in the medium .

SEM analysis

SEM images with magnifications, the silver nanoparticles are agglomerated. In these micrographs it was observed that the nanoparticles were in the size ranging from 20-70 nm with a variety of morphology.

Table 1. UV Visible studies of Nanoparticles

S.No	Name of the fungi	OD Values
1.	<i>A.clavatus</i>	0.562
2.	<i>A.fumigatus</i>	0.570
3.	<i>A. fumiculous</i>	0.578
4.	<i>A.luchensis</i>	0.578
5.	<i>Vericillium longisporum</i>	0.569
6.	<i>A. Oryzae</i>	0.569
7.	<i>P.ferquentans</i>	0.160
8.	<i>Fusriaum oxysporum</i>	0.180

Sawle *et al.*, (2008) synthesized Au-Ag alloy nanoparticles using *Fusarium semitectum* and observed that the band corresponding to the surface plasmon resonance occurs at 545nm for gold nanoparticles and 443 nm for silver nanoparticles. In the present study *Fusarium oxysporum* was mixed with aqueous solution of the silver ion complex and it started to change the colour from pale yellow to dark brown colour(Fig 1) which indicates the formation of silver nanoparticles. It is generally recognized that

UVVis spectroscopy could be used to examine the size and shape controlled nanoparticles in aqueous suspensions (Willey *et al*, 2006). Sawle *et al*, (2008) reported that the peak were at 1643, 1543, 1405, and 1045cm⁻¹ region is a characteristic of proteins and enzymes that have been found responsible for the reduction of metal ions by the fungal mediated synthesis of metal nanoparticles. SEM micrographs have confirmed that silver nanoparticles of 20- 70nm

in the size with various morphology are synthesized have been used for antibiotic assay and act as disinfecting filters and coating materials (Singh *et al* (2008)). This fungus mediated synthesis of silver nanoparticles would be characterized and further investigations are confirmed pertinent to the techniques like calcinations and Energy Dispersive Spectroscopic studies.

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