Review Article

A Review on "Synthesis of Silver Nanoparticles by Biological Approach"

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ABSTRACT

Although there are many Metal Nanoparticles as the part of Nanotechnology, among which Silver Nanoparticles has shown their significance in Diagnostic, Antibacterial, Conductive and Optical applications. This evolved to synthesize the silver Nanoparticles by Physical, Chemical and Biological methods rarely. Toxicity may arise from Silver Nanoparticles synthesized from conventional methods like physical and chemical methods. These techniques use the harmful reducing agents to produce Nanoparticles because, the type of reducing agents used for synthesis of Silver Nanoparticles are crucial for the determination of cytotoxicity. Hence, the green synthesis came into the picture. This paper mainly discusses about advantage in biological way of synthesis which means the synthesis of Silver Nanoparticles by using microbial enzymes and plant secondary metabolites. In Microbes, Enzymes acts as Reducing, Stabilizing and Capping agents where as in plants metabolites and phytochemicals acts as capping and reducing agents. Mostly source of silver is from silver sulphate and silver nitrate. Silver nitrate converts to silver ions when added with microbial and plant extracts. These silver ions utilize the machinery of living cells to develop into elemental form of silver combined to form colloidal Silver Nanoparticles with different shapes like spherical, hexagonal, octagonal, diamond and thin sheets.

Keywords: Nanoparticles, Antibacterial activity, Green synthesis, Reducing agent, Microbial synthesis.

INTRODUCTION

Norio Taniguchi coined the term nanotechnology in the year 1974 were "Nano" means "dwarf" in Greek. Nanotechnology had gained an immense importance in the field of biomedical, [1] drug delivery, [2] healthcare and environment system. [4] Nanoparticles are colloidal particles ranging from 1 nm to 100 nm in nanometer scale these particles are different from bulk materials because of their unique optical, conductive, mechanical, electronic and chemical properties. [5]

Nanoparticles are from many sources like organic, inorganic, magnetic but when compared to other metal Nanoparticles acquired the special importance. Metals such as lead, copper, asbestos, cadmium, [7] aluminium, chromium, manganese, ferrous, gold and silver shown its capability in treating various diseases irrespective of the targets in the body. The silver Nanoparticles also ranges from 1 to 100 nm in size usually as the general Nanoparticle exists. However, metal Nanoparticles has many applications in nanomedicine very potential towards cancerous tissue. [9]

In this review, we discuss the synthesis of silver Nanoparticles by other than frequently used conventional methods like physical and chemical methods (especially by electron irradiation, laser ablation, chemical reduction and electron deposition). Biological methods are less toxic, less time consumed, easily available, eco-friendly.[13]

Biological synthesis of Nanoparticle is the green method of synthesis using either microbes and its enzymes or plants & its secondary metabolites. In microbes, enzymes act like capping, reducing, stabilizing agent. Whereas in plants, metabolites and phytochemicals acts like capping and reducing agents. Type of reducing agents used for synthesis of Nanoparticles are crucial for the determination of cytotoxicity. When both combined separately with metals (like Au, Ag) converts to metal salts which on enzymatic action leads to the formation of metal Nanoparticles further on purification gives the nanoparticle powder. This type of synthesis is a bottom-up technique. The effort has been put forth for the production of silver.

Nanoparticles by microorganisms like Algae, bacteria, fungi & yeast and plant parts like roots, leaves, latex, bark, stem and seeds.

Advantages of biological synthesis are availability of biomaterials, prevention of particle aggregation, provides eco-friendly and pollution free nanoparticle, requires less time to prepare, large variety of active function groups promotes the reduction of silver ions, the solubility of this nanoparticle are high when compared to physical and chemical methods.

MICROBIAL SYNTHESIS OF SILVER NANOPARTICLES

Microorganisms are the 1 millionth the size of the matter. These are called as Nano factories because they lead to synthesis of nanoparticle. Synthesis of nanoparticle by microbes mostly occurs at alkaline pH conditions and temperature also plays an important role in size of silver nanoparticles at room temperature it produces 50 nm size and at more than that leads to decrease in the size. Due to the antimicrobial nature of the silver helps in producing the silver Nanoparticles by microorganisms. The first biologically synthesized silver Nanoparticles intracellularly

within the periplasmic space by Pseudomonas stutzeri AG259, [13] which is a bacterial strain isolated near silver mines in 2000. The Nanoparticles were prepared through reduction of Ag ions with growth cultures in dark conditions. Morphology, size, Ag+ ions reduction can be characterized by TEM (transmission electron microscope), dynamic light scattering and atomic absorption spectroscopy. Silver Nanoparticles are produced with different sources of bacteria, fungi, yeast & algae. Bacterial strains like bacillus *Amyloli quefaciens* (44), *Actinobacter calcoaceticus*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Bacillus licheniformis* and Klebsiella.

General process involved in bacteria:

The process involved in synthesis with microbes include preparation of microbial sample by appropriate incubation conditions to grow culture. The obtained sample is when added with $AgNO_3$. Silver nitrate

converts to Ag+ and NO_3^- ions. Ag+ ions enters the microbial cell which utilizes the nitrate reductase enzyme and stabilizes with one electron from conversion of NADH to NAD+ reaction (Fig.1).

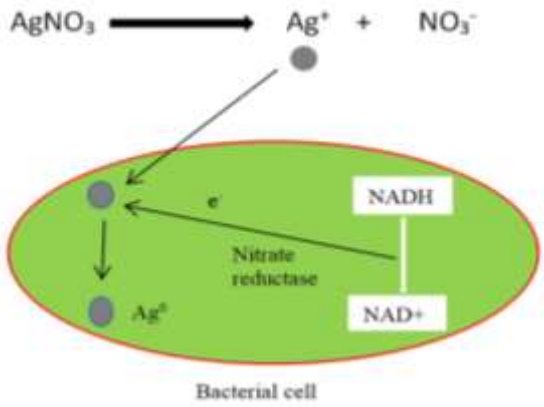

Figure 1: Diagrammatic representation of silver nanoparticles synthesis in bacterial cell

There are several examples excellent in synthesizing silver Nanoparticles such as:

E. coli

Due to its free availability in nature it is cultured by using 3 different media such as Luria-Bertani Lennox medium (LB), Luria-Bertani medium plus nitrate (LBN) and Luria-Bertani medium plus Lactose (LBE) and harvested for synthesis of silver Nanoparticles. The harvested culture is mixed with $AgNO_3$ at concentration of (1% v/v). The reaction occurs between the silver ions and E. Coli. Ag+ ions convert into atomic silver. Hereafter silver Nanoparticles are formed, Which is indicated by change in colour from yellow to reddish brown. [14]

Morganella species RP-42

Isolated from mid gut of an insect. When all the strains of morganella were cultured and incubated under 37°C except morganella psychrotolerans which is under 20°C treated with 5mM of silver nitrate solutions in shaker incubator with 200rpm. This must be carried under dark conditions, leads to the formation of silver nanoparticles within 5days. The Nanoparticles formed extracellularly with plasmon resonance between 400 and 500 nm. [15, 16]

Salmonella typhimurium

Gram negative facultative anaerobe grown on casein media was taken and centrifuged at 5000rpm for 15min. The superficial liquid is used for the synthesis of silver Nanoparticles. Distilled water is used as solvent. 1ml of superficial liquid is added to 100ml of $AgSO_4$ at concentration of 0.0005M. The reaction between two can lead to silver Nanoparticles within few minutes. The color change to brown indicates silver Nanoparticles. Range of approximately 50 to 150 nm Nanoparticles are produced.[17]

Pseudoalteromonas lipolytica

The cells are the marine bacteria obtained from the Yangtze river. Pre-grown cells are exposed to osmotic shock treatment by re-suspending them in distilled water. This was centrifuged and later filtration is done to obtain the supernatant was reacted 4ml of AgNO3 to form silver Nanoparticles. [18, 19]

Synthesis by Actinomycetes

Actinomycetes are facultative anaerobic bacterial forms the colonies which seems fungi like branched network of hyphae. The cultures of actinomycetes was incubated at 27°Cand agitated at 220rpm which is harvested after 5 days and centrifuged at 12000rpm for 10min. This sample was taken added to silver nitrate of 0.1M concentration. The sample contents act as reducing agents. The reaction between sample and silver nitrate helps to form silver atoms combined to giver colloidal Silver Nanoparticles. [20]

Synthesis by yeast

Yeast is utilized to produce the Silver Nanoparticle where metal salts are combined with enzymes released from yeast. The yeast must be stored & cultured in the yeast peptone dextrose medium with 1% yeast extract, 2% peptone, 2% dextrose. The cultured yeast incubated at 30°C 9-10 hrs then added to silver nitrate. [21]

Synthesis by fungal species

The various studies stated that silver Nanoparticles produced by fungal species are stable, overriding, uniform size and shape in dispersed phase. Even fungus releases some proteins like enzymes acts as reducing agents does interaction with metal ions.[22] This strain produces nanoparticles both intracellularly and extracellularly were metal ions dragged on the surface of the cell and reduces in presence of proteins (enzymes). Many fungal species such as volvariella volvaceae and fusarium oxyporum, [23] helps in producing combination of gold and silver Nanoparticles.

For example, verticillium fungus gives the intracellular synthesis of silver Nanoparticles of 25 nm (Table No.1).

Table 1: Some of the examples of fungal species used for biosynthesis of silver nanoparticles

Fungal species	Production site	Size
Fusarium Oxyporum	Extracellular	5-15nm [23]
Aspergillus Fumigatus	Extracellular	5-25 nm [24]
Coriolis versicolor	Intracellular	400 nm [25]

PLANTS UTILIZED FOR SYNTHESIS OF SILVER NANOPARTICLES

Plants & its parts such as roots, leaves, stem, bark all these contain the carbohydrates, pigments, proteins, nucleic acids and various other secondary metabolites which acts as the reducing agent to produce silver Nanoparticles. By the use silver Nanoparticles from plant extracts shown bactericidal action towards both Gram-positive and Gram-negative bacteria.

General process

In this process synthesis of silver Nanoparticles by plant extract containing bio molecules reacted with the $AgNO_3$ (metal ionic solution) converts to silver ions. [26] Further these silver ions surrounded by bio molecules gives rise to nucleation and accumulation of capping agents & stabilizing agents results in silver Nanoparticles. Key factors which influence this are pH, concentration, reaction time, reaction temperature. Plant extracts helps in producing various nanoparticles such as quantum dots, gold & silver. (Fig.2) (Table No.2).

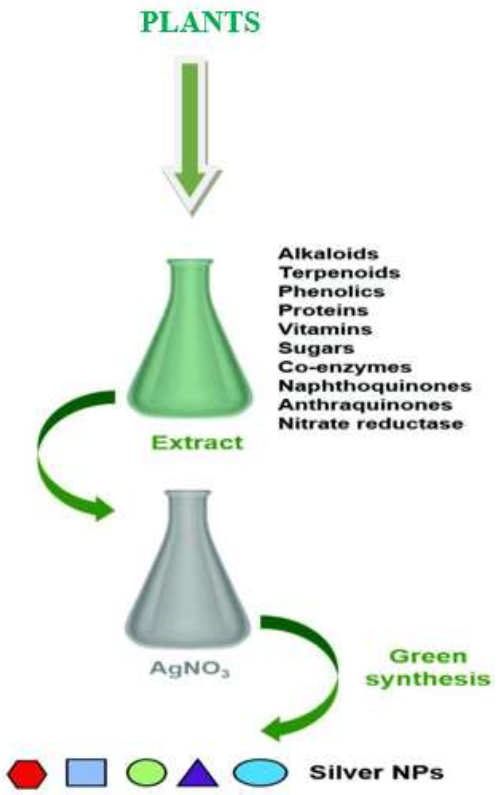

Figure 2: Bio-synthesis of Silver Nanoparticles by Plants

Table 2: There are many examples under clinical study obtained from different sources.

Plant source	Plant Part used	Time consumed	Characterization by
Azadiractha indica (neem)	Leaf extract	24 hrs	TEM, DSL Analysis, UV Spectrophotometer. [27]
Amomum villosum (cardamom)	Dried fruit extract	-	X-ray diffraction, DSL, FTIR, FE-TEM, Selected area electron diffraction. [28]
Coriandrum sativum	Leaf extract	1 hr	UV, X-RAYS, TEM, FTIR. [29]
Cinnamon	Bark extract	8 hrs	X-RAYS, FTIR, UV Spectrophotometer, FESEM, EDAX. [30]
Camellia sinensis	Leaf extract	-	UV, FTIR, TEM. [31]
Ginkgo biloba	Leaf extract	15-100min	UV, FTIR, X-RAYS, Energy dispersive x-ray spectroscopy, Field Emission SEM. [32]
Persimmon (Diopyros kaki)	Leaf extract	-	UV, TEM. [34]
Platanus orientalis	Leaf extract	2 to 100min	UV, FTIR, TEM. [34]
Tectona grandis (Teak)	Seed extract	1hr	FTIR, XRD, UV, SEM. [35]

CONCLUSION

The silver Nanoparticles have many properties applicable to threaten diseases in humans and animals due to its potential efficacy which are under clinical investigation studies. The use is not only in field of agriculture, but also in technological improvement.

Biological approach is an alternative method in order to reduce toxicity offered by physical and chemical methods for fabrication of silver Nanoparticles. As they are having efficient solubility, stability, distribution, and absorption capacity into the cells. Due to its antimicrobial properties they are used as pathogen killing agent. The studies are going on to know the long term and prolonged effects on human cell.

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