ABSTRACT: Over the few decades, the interest of scientists in the field of nanotechnology has increased. The production of metallic nanoparticles is an important and active area of interest. Although numerous physical and chemical methods of nanoparticle synthesis are available, it is more convenient and helpful to use biological systems, such as micro-organisms (Bacteria, fungi) for nanoparticle synthesis. This system gives an alternative green way of synthesizing low-cost, nontoxic metal nanoparticles. This review provides an overview of useful microorganisms in the synthesis of metallic nanoparticles and also highlights the potential importance of the living systems in various fields.

KEYWORDS: Nanotechnology, metallic nanoparticles, micro-organisms, bacteria, fungi.

I. INTRODUCTION

In recent years, the topic of nanoparticles has received a great attention. The term ‘nano’ is a Greek word meaning “Dwarf “, which denoted the measurement on the scale of one billionth ($10^{-9}$) of a meter in size (29, 39). Nanoparticles are defined as particulate dispersion of solid particles with at least one dimension at a size range of 10-1000 nm (25, 39). A size of a typical virus is around 100 nm wide and that of DNA strand is 2.5nm in diameter (23). The most important feature of nanoparticles is their surface area to volume ratio, allowing them to interact with other particles easily (40). Different physical, chemical and biological methods have been employed to obtain the metallic nanoparticles with different properties. However, physical and chemical methods have certain drawbacks like use of toxic chemicals, high energy requirements etc. In contrast, a biological method seems to be promising way of nanoparticle synthesis. The synthesis and assembly of nanoparticles would benefit from the development of clean, nontoxic and environmentally acceptable “green chemistry” procedures, probably involving organisms ranging from bacteria to fungi and even plants (5, 34). 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 nano-materials. Hence, unicellular as well as multicellular organisms such as bacteria, yeast, viruses, fungi, algae and also the plants are well known to produce nanoparticles either intracellularly or extracellularly. Nanoparticles have diverse possible area of applications such as biotechnology, electronics, pharmaceuticals, biomedical and environmental sciences.

II. NANOPARTICLE SYNTHESIS BY FUNGI

Production of metal nanoparticles by using fungi is referred as mycofabrication. Fungal biosynthesis of nanoparticles has emerged as “Bio-nano-factories” in recent years. They synthesize the enzymes which help in reduction of metal ions to nanoparticles and also favor the large scale production of different metal nanoparticles such as Gold, silver, iron, cadmium, platinum, magnetite etc. (27). Fungi can accumulate metal ions by physico-chemical and biological mechanisms including extracellular binding by metabolites and polymers, binding to accumulation (26). This gives an ease in the downstream processing and also in the handling of fungal biomass. Another advantage of using fungi mediated green synthesis of metal nanoparticle includes the rapid and eco-friendly approach.
Verticillium sp. was used to obtain nanoparticles with well-defined dimensions and good mono-dispersion. Mukharjee et al. (2001) studied synthesis of gold nanoparticles with well-defined dimensions using Verticillium sp. These results have documented that the trapping of AuCl₄⁻ ions on the surface of fungal cells could occur by electrostatic interaction with positively charged groups (such as lysine residues) in enzymes that are present in the cell wall of the mycelia(26). Fusarium oxysporum strains were also used in the extracellular synthesis of silver nanoparticles or silver hydrosols (1). A nitrate dependent reductase and shuttle quinines from F.oxysporum are found to be responsible for the synthesis of silver nanoparticles (12).

Bhainsa et al. (2006) demonstrated a rapid synthesis of fairly monodispersed silver nanoparticles by using filamentous fungus Aspergillus fumigatus within 10 minutes (3). Platinum nanoparticles were synthesized by using a fungus Neurospora crassa. Castro-Longoria et al.(2012) was able to synthesize platinum nanoparticles of size 4-35 nm intracellularly by using N.crassa (6). Trichoderma asperellum and Trichoderma reesei are able to produce silver nanoparticles when exposed to silver salts (27, 31).

III. NANOPARTICLE SYNTHESIS BY BACTERIA

Among all the natural resources, prokaryotic bacteria have been heavily researched for the synthesis of metallic nanoparticles. Due to their relative ease of manipulation and ability to adapt to extreme conditions, bacteria are preferred to be a good choice for the nanoparticle synthesis. Another reason for selecting bacteria is they are fast growing and easy to cultivate and also their culture conditions such as temperature, pH, incubation time can be easily controlled, which plays an important role in the production of nanoparticles. He et al. (2006) found that change in the P² of the growth medium results in the production of nanoparticles with different sizes and shapes(15). Slawson et al. (1992) discovered that the bacterial strain of Pseudomonas stutzeri AG 259 isolated from silver mines, produced silver nanoparticles as they accumulated Ag nano particles, within their periplasmic place (38). Klaus et al. (1999) observed that when P. stutzeri AG 259 was treated with concentrated aqueous solution, they were able to produce particles of larger size(200 nm)(20).

Husseiny et al. (2007) used a cell filtrate of Pseudomonas aeruginosa for the biosynthesis of extracellular gold nanoparticles by reduction of gold ions. The use of cell filtrate helped in achieving the production of controlled sized and polydispersed nanoparticles (17). Nair and Pradeep (2002) reported that common Lactobacillus strains present in buttermilk produces microscopic gold, silver and gold-silver alloy crystals of well-defined morphology when exposed to high concentration of metal ions. These Lactobacillus strains produces nanoparticles intracellularly and are also able to maintain their cell viability even after the growth of crystals inside the cells (28).

Morganella morganii produces pure elemental Cu nanoparticles. M.morganii uptakes Cu ions when treated with Cu solution and this ions binds to either metal ion reductase or similar proteins. Reduction of the ions to metallic Cu⁰ occurs which then accumulates extracellularly as nanoparticles once eﬄuxed out of the cell(32). Konishi et al. (2004) achieved a gold nanoparticle by using mesophilic bacterium Shewanella algae, by varying pH conditions (21). Magnetotactic bacteria were also used for the synthesis of magnetic nanoparticles (33). Actinobacter sp. is able to synthesize magnetic nanoparticles when treated with suitable aqueous iron precursors under fully aerobic conditions (4).

IV. NANOPARTICLE SYNTHESIS BY YEAST

Few studies are reported about the synthesis of metallic nanoparticles by yeasts. Dameron et al.(1989) conducted biosynthesis of Cadmium Sulphide (CdS) nanocrystals by using two yeast-Candida glabrata and Schizosaccharomyces pombe. The nanocrystals produced by this yeasts are used in quantum semiconductor crystallites (8). Another yeast strain MKY3, a silver tolerant species is used for the synthesis of silver nanoparticles. Kowshik et al.(2003) demonstrated a study, in which MKY3 was able to produce a silver nanoparticles of size ranging from 2-5 nm. In this study MKY3 was challenged with silver solution in the log phase of growth. This study also revealed that large quantities of silver nanoparticles could be synthesized by using MKY3 strain by using standardized conditions (22).
Very few reports are available, in which algae are used as a source for metallic nanoparticle synthesis. Singaravelu et al. (2007) first reported the use of marine algae in metallic nanoparticle synthesis. Marine algae Sargassum wightii was able to synthesize highly stable gold nanoparticles extracellularly(37).

VI. NANOPARTICLE SYNTHESIS BY VIRUSES

Use of viruses for the synthesis of nanoparticle is very less studied and hence very few reports are available. Douglas et al. (2002) have reported that cowpea chlorotic mottle virus and cowpea mosaic virus can serve as a nucleation template for mineralization of inorganic materials(10,11), but the exact mechanism is not known. Study conducted by Shenton et al.(1999) revealed the tobacco mosaic virus was able to synthesize lead sulphide (PbS) and Cadmium sulphide(CdS) crystalline nanowires (36).

VII. NANOPARTICLE SYNTHESIS BY PLANTS

From last few decades research on the green synthesis of nanoparticles by using plants or plants extracts has been increased a lot (30). The use of plants and plant extracts is giving promising results in metallic nanoparticle synthesis. Many plants have been studied for the synthesis of nanoparticles. The production of spherical silver nanoparticles of size 3-20nm was observed when the leaf extract of Ocimum sanctum was treated with 1mM AgNO3 solution (24). Another study done by Shankar et al.(2004) showed a single step reduction of AuCl4 ions to produce triangular shaped gold nanoparticles by using a plant extract of lemongrass Cymbopogon flexuosus (35). An intresting investigation documented by Gardea- Torresdey et al.(2003) showed a synthesis of quantum dots by using Alfalfa roots. The roots absorbs the Ag ions from the medium when traeted with solution and transfers to the shoot. In the shoot a special arrangement of Ag atoms occurs where the silver nanoparticles are formed by joining the Ag atoms (14). Armendariz et al. (2004) reported a pH dependent gold nanoparticle synthesis by the plant Avena sativa (Oat). The Au (III) ions binds to the biomass and depending on the pH, Au nanoparticles of variable sizes are formed. In this study, it was observed, at low pH, larger size nanoparticles are formed. At pH 2 nanoparticles of size 25-85nm were synthesized, while at pH 3 and 4, nanoparticles of 5-20nm were synthesized. The variability in the size is due to the aggregation process of gold nanoparticles is favored over nucleation process at low pH. Whereas at pH 3 and 4, more functional groups allows the binding of gold nanoparticles to biomass, favouring the more synthesis of nanoparticles(2). Another plant showing the potential for metallic nanoparticle synthesis is Cinnamomum camphora. The leaf extract produces a gold as well as silver nanoparticle with controlled shape. The shape control is attributed to the protective and reductive biomolecules produced by the plants (16).

VIII. APPLICATION OF NANOPARTICLES

Nanotechnology has received a great attention of biologist because of their wide range of applications in different fields. Few applications of nanoparticles produced by micro-organisms are:-

- **Regulation of biological processes:-**
  By using suitable nanoparticles, different biological processes can be regulated where nanoparticles can be used to enhance the gene expression in the cell; in which hybrid polymer-protein conjugate nanoparticles are useful which increases the delivery of the DNA to the cell nuclei. Silver nanoparticles have been used in the treatment of cancer, where a complex of antibody and a nanoparticle is used which targets a cancer cell to create a free radical which would then kill the affected cells(9).

- **Antimicrobial agents:-**
  Gold and silver nanoparticles are used as an antimicrobial agent, as those are showing antibacterial effect against many pathogenic microorganisms. These nanoparticles have the ability to create reactive oxygen species which causes irreversible damage to the bacteria and also have a strong affinity in binding to DNA or RNA which interferes with the microbial replication process(9).
Copper nanoparticles are used as an antimicrobial agent, as they exhibit activity against many disease causing microorganisms (41).

- **Intercalating agents:**
  Silver nanoparticles have a large number of applications such as intercalation materials for electrical batteries. It is also used as an optical receptors, which is useful in many chemical reactions where it acts as a catalyst and also in many healthcare devices as it poses an antibacterial properties.(12).

- **Catalytic agent:**
  Platinum nanoparticles are also used as a catalyst in many chemical reactions because of its superior catalytic activity. It is also used in the fuel cell technology, where it acts as a cathode as well as anode, as it shows an effective redox reaction (7).

- **Other applications:**
  Magnetite (Fe₃O₄) and siliceous material produced by different bacteria and diatoms are used in optical coating for solar energy applications and as ion insertion materials for electrical battery application (18).

Nanoparticles are also useful in the bioremediation of radioactive wastes generated by nuclear power plants. Certain strains of bacteria have the special capabilities for the clean-up of uranium contaminated waste waters (13).

### VIII. CONCLUSION

Biological systems have a great potential to provide a sustainable, resource efficient and cheap method for the production of nanoparticles with a wide applications. By using the micro-organisms such as bacteria, fungi, yeast and molds nanoparticles with different shapes and sizes can be obtained. Chemical and physical methods used for the production of nanoparticles involves a toxic chemicals and are also harmful to the environment, whereas theseliving organisms provides a green pathway for the production of metallic nanoparticles such as gold, silver, platinum, copper, iron, palladium etc. However, the field of biological production of metal nanoparticles using living system is relatively new and unexplored. Hence, it shows great opportunities for budding biologists to study the biological systems for the production of metal nanoparticles where many different aspects of these biological systems can be studied and later can be emerged for commercialization purposes.

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