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Review Article

Nanoparticle Formulation Nanoparticles, Gold (Au) Nanoparticles, Copper (Cu) Nanoparticle, Nanoscale Zero Valent Iron and Advancing Algal Nanoparticle Synthesis in Nano Biotechnology

Amira Imran Hussien[1*]

[1]Al-Qasim Green University, College of Biotechnology Iraq

***Corresponding Author:** Amira Imran Hussien
Al-Qasim Green University, College of Biotechnology Iraq

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Abstract: There is a rising demand for biocompatible and ecologically friendly nanomaterials, which has prompted the development of nano-biotechnology as a bridge between the two fields. When compared to more traditional physical and chemical approaches, green synthesis for nanoparticle creation is clearly the superior choice. This method reduces the need of harmful chemicals and severe reaction conditions by making use of naturally occurring reducing and stabilising agents, such as plants, bacteria, fungi, algae, yeast, and other microbes. Common characterisation methods, the processes involved in nanoparticle production, and the impact of biological components on particle size, shape, and stability are covered. A literature review was conducted using well-known scientific databases such as PubMed, ScienceDirect, Scopus, Springer, Wiley, and Taylor & Francis. Paying close attention to the trends and limitations in reported performance, the functional applications of green-synthesized nanoparticles in the biological, agricultural, catalytic, and environmental domains are assessed. Furthermore, within the framework of environmental remediation, the photocatalytic and pollutant degrading capabilities of nanoparticles that are biologically produced are emphasised. One way to overcome the limitations of chemical and physical procedures is to use biological resources for green synthesis of silver nanoparticles (AgNPs). This method is the most straightforward, cost-effective, quick, and eco-friendly option. A wide variety of biological resources, including plants and their many components, bacteria, fungi, algae, and so on, could be employed in the environmentally friendly production of bioactive AgNPs. A number of environmentally friendly methods for the quick, easy, and non-toxic production of AgNPs from biological resources have been published in the last several years. During biosynthesis, plant extracts contain a variety of biomolecules that serve as reducing and capping agents. These biomolecules include flavonoids, terpenoids, alkaloids, phenolic compounds, and vitamins. Similarly, various microbes produce metabolites, both primary and secondary, which are essential reducing and capping agents in the synthesis process. Researchers are very interested in biosynthesized AgNPs due to their potential uses in various biomedical disciplines. The bactericidal activity of AgNPs is its most widely used property. Researchers are looking at the medicinal potential of AgNPs as possible antibacterial agents because of the rise of germs that are resistant to many drugs. Several studies have shown that biosynthesized AgNPs are highly effective against a wide variety of human diseases. The tiny size and enormous surface area of AgNPs make them highly effective against bacteria. They can enter bacterial cells, harm their membranes, create reactive oxygen species, disrupt DNA replication and protein synthesis, and ultimately kill the bacteria.

Keywords: Nanoparticle, Gold (Au) Nanoparticles, Copper (Cu), Zero-Valent Iron, Nano-Biotechnology.

INTRODUCTION

The advancement of functional nanomaterials for use in biology, agriculture, and medicine has propelled nanoscience and nanotechnology to the forefront of the materials research revolution. Improving the efficiency, security, and longevity of biological systems is the goal of nanobiotechnology, a subfield of this larger field that combines nanoscale materials with systems biology. Along with these advancements, there are still a number of drawbacks associated with the

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conventional physical and chemical methods of synthesising nanoparticles, such as the need for large amounts of energy, the use of harmful reducing agents, and the creation of harmful by-products that pose a threat to both human and environmental health [1,2]. As a potential eco-friendly method for synthesising nanoparticles, "green nanotechnology" has gained popularity as a potential answer to these needs. Plants, bacteria, fungus, and algae are examples of naturally occurring reducing and stabilising agents that provide the basis of the principle. Benefits such as internal biocompatibility, minimal toxicity, and low reaction conditions are clearly outlined for these biological systems. A wide variety of phytochemicals, including flavonoids, phenolics, terpenoids, and proteins with a dual action (i.e., reducing metal ions and stabilising nanoparticles), are found in plant extracts, according to recent studies. Just as bacteria produce enzymes and metabolites, which enable the regulated creation of nanoparticles in terms of size and structure, so do microorganisms [3]. Nanomaterials produced by living organisms have discovered uses beyond the medical industry. For agricultural use, green-synthesized nanoparticles have shown promise as nanofertilizers and nano-pesticides, thanks to their ability to transport nutrients, promote nutrient development, and lessen the chemical burden on soil and water. They have proven useful in environmental science for both sensing and pollution treatment. Nanoparticles generated from microbes and plants are being used more and more in diagnostics and medicine for antimicrobial treatment, medication administration, wound healing, and medical imaging because of their many useful properties and low risk of side effects [4] . Research into nanotechnology is just getting underway, but it has the potential to revolutionise many areas of science and technology, particularly in the creation of new nanomaterials and nanoparticles. Biomedicine, agriculture, pharmaceutics, textiles, food technology, catalysis, sensors, mechanics, electronics, and optics are just a few of the many scientific domains that have taken an interest in nanoparticles (NPs), which are very tiny particles ranging in size from 1 nm to 100 nm. Nanoparticles come in a wide variety of forms, each with its own set of properties; examples include silver, gold, zinc, cadmium sulphide, copper, iron, titanium dioxide, and many more [5]. Due to their extensive use in various biomedical fields as anti-cancer, anti-inflammatory, anti-fungal, antioxidant, drug delivery, wound dressings, biosensors, biocatalysis, etc., silver nanoparticles (AgNPs) have risen to prominence as a popular nanoparticle research topic in recent decades. Recent research has demonstrated that AgNPs produced in a sustainable manner have potent antibacterial, antioxidant, and anti-cancer properties [6-8]. Biosynthesized AgNPs were also found to be effective in degrading a range of harmful compounds [9]. Water filtering, sanitization, food preservation, cosmetics manufacture, nano-insecticides, nanopesticides, etc. are only a few of the many biotechnological uses for green-synthesized AgNPs [6]. Several Gram-positive and Gram-negative harmful bacteria have shown interest in green synthesised AgNPs as a possible antibacterial agent. These nanoparticles are often synthesised using a variety of physical and chemical techniques, including physiochemical, electrochemical, photochemical, chemical reduction, and microwave irradiation. Due to the utilisation of poisonous substances, expensive and labor-intensive equipment, and the development of hazardous byproducts, these approaches are expensive and hazardous [7]. Researchers are increasingly looking to biological ways for the facile, economical, environmentally friendly, and non-toxic creation of nanoparticles while physicochemical methods have several limitations. Instead than using costly and harmful chemicals, green synthesis makes efficient use of natural substances to reduce, cap, and stabilise. Plants (and all their parts), bacteria, fungus, algae, and other biological resources could be used for the green synthesis of bioactive nanoparticles [8]. Antimicrobial activity and green production of AgNPs utilising microorganisms or plant extracts have recently been the subject of extensive research. Polymers 2021, 13 x 2, containing 24 distinct nanoparticle kinds, each with its own set of properties; these include, but are not limited to, silver, gold, zinc, cadmium sulphide, copper, iron, titanium dioxide, etc. Due to their extensive use in various biomedical fields as anti-cancer, anti-inflammatory, anti-fungal, antioxidant, drug delivery, wound dressings, biosensors, biocatalysis, etc., silver nanoparticles (AgNPs) have risen to prominence as a popular nanoparticle research topic in recent decades. Recent research has demonstrated that AgNPs produced in a sustainable manner have potent antibacterial, antioxidant, and anti-cancer effects. Biosynthesized AgNPs were also found to be effective in degrading a range of harmful compounds [9]. Water filtering, sanitization, food preservation, cosmetics manufacture, nano-insecticides, nanopesticides, etc. are just a few of the many biotechnological uses for green-synthesized AgNPs. These nanoparticles are often synthesised using a variety of physical and chemical techniques, including physiochemical, electrochemical, photochemical, chemical reduction, and microwave irradiation. Because of the poisonous substances used, the expensive and labor-intensive equipment required, and the harmful byproducts generated, these approaches are both expensive and hazardous [10, 11]. Researchers are increasingly looking to biological ways for the facile, economical, environmentally friendly, and non-toxic creation of nanoparticles while physicochemical methods have several limitations. Instead than using costly and harmful chemicals, green synthesis makes efficient use of natural substances to reduce, cap, and stabilise. One potential source for the environmentally friendly production of bioactive nanoparticles includes a wide variety of plants and their many components, including roots, leaves, fruit, etc., as well as bacteria, fungi, algae, and other similar organisms [12]. There has been recent progress in the green production of AgNPs by leveraging the antibacterial activity of microorganisms or plant extracts.

Nanoparticle formulation using microbes and plant sources

Because it is a straightforward, environmentally friendly, and non-toxic approach that does not include harmful chemical reagents, the green synthesis of nanoparticles using plant extract and microorganisms has lately garnered a lot of attention. The creation of nanoparticles can be achieved in controlled conditions under mild reaction conditions thanks to these biological systems, which act as stabilising and reducing systems [13]. Leaf, root, stem, and fruit extracts derived

from plants can be utilised in the creation of nanoparticles in plants, whether the extraction method is aqueous or hydroalcoholic. Polyphenols, flavonoids, tannins, terpenoids, and alkaloids are just a few examples of phytochemicals. These molecules help stabilise surfaces and reduce metal ions to smaller sizes. There is a close relationship between the size, morphology or shape, and stability of nanoparticles and the composition of plant extracts, reaction pH, temperature, concentration of metal ions, and incubation period. There is evidence that plant extracts rich in biopolymers can improve the biomedical applications of nanoparticles by increasing their colloidal stability and functional performance [14]. Some microorganisms are able to synthesise nanoparticles both inside and outside of their cells; they include yeast, bacteria, and fungi. These systems employ reductases and other enzymes, as well as proteins and polysaccharides found in the microbial cell wall or released into the culture medium, to stabilise nanoparticles and improve metal ion reduction. Due to downstream processing and higher scalability, extracellular synthesis is often preferred. Culture factors that substantially impact yield and physicochemical characteristics include growing medium composition, incubation temperature, pH, and concentration of metal precursor. Therapeutic and antibacterial uses benefit from the improved control over surface functions and particle consistency in microbial production. In general, nanoparticle formulation mediated by plants and microorganisms offers a sustainable alternative to traditional methods of nanoparticle synthesis. The versatility of these technologies makes them ideal for use in pharmaceutical and biomedical applications, as they enable the creation of biocompatible nanoparticles with tuneable features [15].

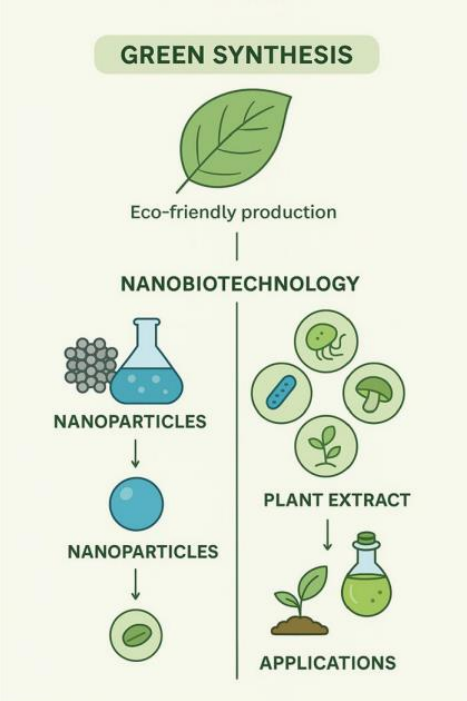

Figure 1: An illustration of the green synthesis of nanoparticles by the use of microbes and plant extracts. Nanoparticles can be formed in mild circumstances using biological sources as reducing and stabilizing agents. These nanoparticles have uses in the biomedical, environmental, and agricultural domains

Gold (Au) Nanoparticles

Nanoparticles of gold (Au) have attracted a lot of attention due to their unique optical absorption capabilities, which are influenced by their size, shape, and surface features. The transportation of large macromolecules, such as proteins and nucleic acids, is one of the many appealing biological uses for these noble nanoparticles. They also provide a secure pathway for the transfer of genetic material and pharmaceuticals, which is essential in the fight against cancer. Although conventional approaches have long been used, new evidence shows that green synthesis technologies are much more effective. These methods simplify characterisation operations, cut down on failure possibilities, and lower costs [16]. Green nanoparticles are attracting attention from throughout the world due to their biocompatibility and many possible uses, including as catalysts, antimicrobials, biofuel producers, and cancer/gene treatment tools. The shape, size, content, volume/surface area ratio, and purity of the constituents give nano-materials their unique features. There are a number of ways to synthesise nanoparticles, but the physical and chemical procedures are very expensive and produce harmful waste. Building nanoparticles from tiny parts (bottom-up) or breaking them down from larger items (top-down) are the two basic approaches to manufacture them. Better, more eco-friendly procedures have recently been discovered by scientists (green synthesis). Aligning with sustainable methods, green synthesis guaranties the absence of harmful chemicals, minimises manufacturing, and utilises ecologically favourable substances [17]. In numerous fields, including as medicine, sensing, and catalysis, gold (Au) nanoparticles are essential due to their exceptional electrical, optical, and catalytic properties. The usage of harmful chemicals and energy-intensive processes is common in traditional AuNP synthesis methods, which raises

environmental issues. However, in order to reduce the environmental impact of producing nanoparticles, green synthesis makes use of reducing agents that are biogenic or derived from plants.

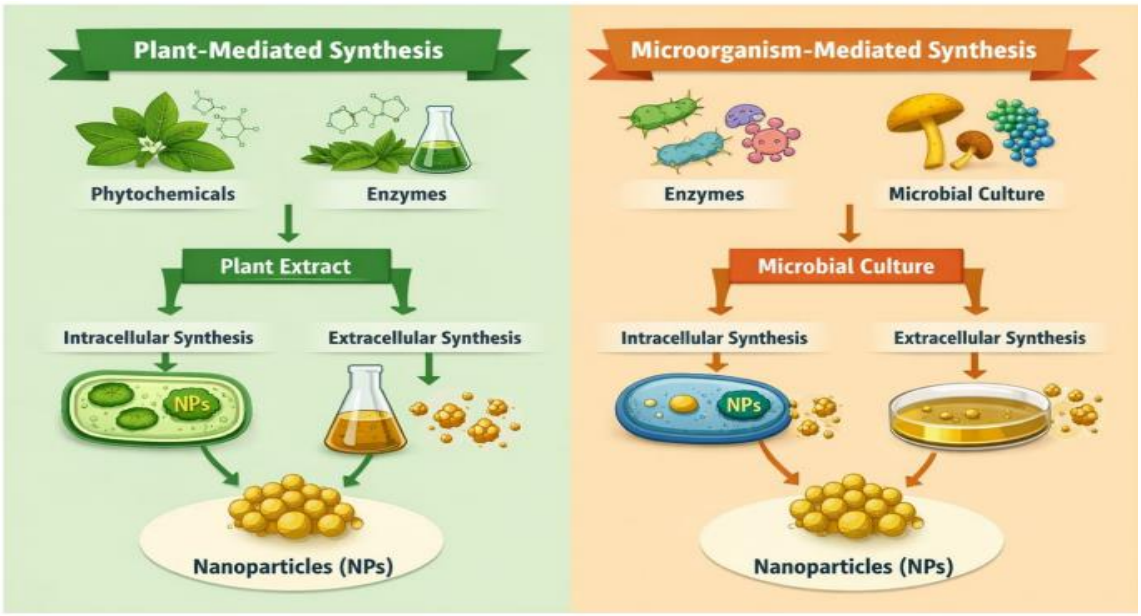

Figure 2: Analyzing the processes involved in the creation of nanoparticles mediated by plants and microbes. Microbes mediate the enzymatic and metabolic pathways that lead to the creation of nanoparticles, either within or outside of cells, whereas plant extracts supply phytochemicals like flavonoids and polyphenols that reduce metal ions

Sustainable Synthesis Method

Choosing an Environmentally Friendly Deterrent: Choose a biocompatible reducing agent with caution; that's the first stage. Natural substances with inherent reducing capabilities are often preferred, such as microbial enzymes, plant extracts, or similar substances. To create nanoparticles out of gold ions, other substances have demonstrated potential, such as aloe vera gel, green tea extract, and microbial enzymes produced by bacteria or fungi [18]. How a Gold Precursor Solution Is Formed: A precursor solution containing gold ions can be prepared by dissolving a gold salt in the right solvent. Chloroauric acid (HAuCl4) is the typical name for this. The concentration of the gold salt and the solvent selection largely determine the size and stability of the resultant NP. The reaction mixture is prepared by combining the gold precursor solution with the environmentally friendly reducing agent that has been chosen. In order to create stable nanoparticles, the reaction mixture is incubated under carefully controlled conditions, including pH and temperature, that encourage the reduction of Au ions. Analysing Nanoparticles: The nanoparticles are studied by microscopy, spectroscopic, and scattering methods. These findings clarify the arrangement and physical properties of AuNPs, including their size and form [19]. In order to make the green synthesis process as efficient as possible, it is necessary to fine-tune the synthesis parameters. These parameters include temperature, pH, and concentration ratios. The goal of this stage is to improve the stability of nanoparticles and gain control over their physicochemical properties. As an alternative to conventional procedures, green synthesis of AuNPs is more sustainable and less harmful to the environment. This method reduces environmental consequences and paves the way for many biomedical, sensing, and catalytic uses by tapping into the reducing power of naturally occurring chemicals. The use of environmentally friendly synthesis processes in the creation of nanomaterials is becoming more and more important as nanotechnology progresses.

Copper (Cu) nanoparticle

One approach to reducing the environmental impact of conventional methods is the "green synthesis of nanoparticles." This involves employing sustainable and environmentally friendly processes to produce nanoscale materials. Several green synthesis techniques have been studied in the context of copper nanoparticles. As a reducing and stabilising agent, plant extracts are a well-known technique. The bioactive chemicals in copper salts are usually sourced from plant extracts combined with an aqueous solution. Copper ions were reduced to generate Cu nanoparticles by use of the extract's reducing agents. Another important factor in nanoparticle stabilisation and agglomeration prevention is the phytochemicals found in the plant extract. In order to maximise the efficiency of the synthesis process, we also meticulously controlled the reaction parameters, including the pH and temperature. In comparison to conventional approaches, this environmentally friendly approach has benefits including lower energy use, biocompatibility, and cost-effectiveness. In addition, plant extracts include numerous bioactive chemicals, which can enhance the copper nanoparticles' functionality. This makes them more versatile and applicable to fields such as biomedicine, sensing, and catalysis.

Nanoscale zero-valent iron (NZVI) (iron and iron oxide NPs)

Iron oxide NPs, NZVI, and Fe NPs are examples of iron-based nanoparticles that can be synthesised in an environmentally responsible way by reducing the usage of potentially harmful chemicals. The use of plant extracts is one typical

method. Another method that can be used to synthesise NZVI is the reduction of zero-valent iron precursors in the presence of plant extracts. This process is similar to that which is used to synthesise Fe NPs, where an aqueous solution containing iron salts is mixed with a plant extract, resulting in the formation of nanoparticles. The controlled oxidation of NZVI or the use of plant extracts with inherent oxidative characteristics can be employed to create Fe oxide nanoparticles. To stabilise the resultant nanoparticles and avoid agglomeration, plant extracts were crucial in the reduction of metal ions. This green synthesis approach is a sustainable and eco-friendly substitute for conventional synthetic pathways due to its many benefits, including low toxicity, cheap cost, and scalability. For synthesis to be successful, each combination of plant extract and iron compound may necessitate somewhat different settings (conditions and parameters). The significance of optimisation in achieving the required nanoparticle characteristics is shown by this, which emphasises the need for changes based on unique materials [20]. Nanoparticles have unique physicochemical properties that make them applicable to many fields. Traditional approaches to synthesising nanoparticles frequently necessitate the use of potentially harmful chemicals and substantial amounts of energy. To the contrary, microorganisms like algae, bacteria, and fungi are utilised in green synthesis to lessen the environmental impact and guaranty sustainability.

Microbes that are resistant to multiple drugs

Because they can cause a variety of infectious diseases, some of which can be fatal, multidrug-resistant bacteria pose a major risk to public health around the world. The overuse of medications, along with mutations, pollution, and shifting environmental circumstances, is leading to an alarming rise in the number of bacterial strains that are resistant to many drugs. Scientists are attempting to find a solution by creating new medications to treat these types of microbial diseases. These bacterial strains are resistant to multiple drugs, but they can be controlled by green synthesised AgNPs.

Green Synthesis of AgNPs

The use of various biological agents in the green synthesis of AgNPs eliminates the production of harmful byproducts while being cost-effective, easy, and environmentally benign. The environmentally friendly production of AgNPs has recently been the subject of much research into both plant and microbial sources.

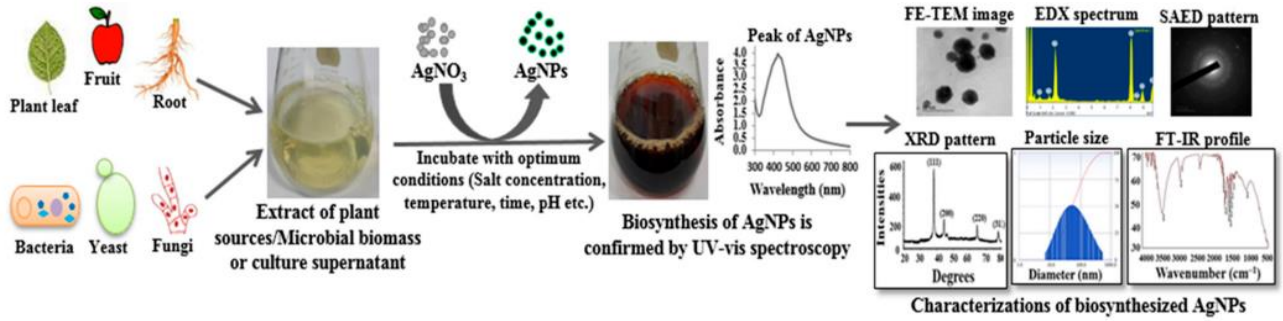

Figure 3: Green synthesis and characterization of AgNPs schematically illustrated

Silver Nanoparticles Produced by Plants

Many plants are readily available, and they are easy and safe to use, hence plant-mediated synthesis of AgNPs is a popular strategy. The green synthesis of bioactive AgNPs has been effectively carried out using various plant parts, such as fruits, roots, flowers, leaves, peels, etc. Alkaloids, flavonoids, terpenoids, tannins, saccharides, phenols, vitamins, enzymes, amino acids, and proteins are only a few of the bioactive substances found in plant extracts [21]. It is easier and more stable to synthesise bioactive AgNPs using plants because these biomolecules are already present in plant extracts. The green synthesis of bioactive AgNPs from various plant components, including fruits, seeds, roots, flowers, stems, leaves, peels, etc., has been the subject of extensive research in recent years. One example is the simple, quick, and environmentally safe production of bioactive AgNPs using an extract from the leaves of Clerodendrum viscosum. They also looked into how well biosynthesized AgNPs worked as antimicrobials against different kinds of harmful bacteria. For the environmentally friendly production of AgNPs, the seeds of Durio zibethinus and the roots of Rheum palmatum were utilised, respectively. In order to synthesise bioactive AgNPs, researchers looked at vegetable peel extracts from a variety of plants, including Cucumis sativus, Lagenaria siceraria, Luffa cylindrica, Solanum lycopersicum, and Solanum melongena. The amount of time it takes to synthesise AgNPs, the size and form of those nanoparticles, and their bioactivity can vary substantially depending on the plant or plant component that was utilised. As an example, AgNPs ranging in size from 10 to 30 nm were produced by reacting Panax ginseng root extract for two hours [22]. In contrast, AgNPs ranging from 5 to 15 nm in size were produced utilising Panax ginseng leaf extract in just 45 minutes after the reaction began. Spondias mombin leaf extract was found to yield AgNPs with a rod or triangle shape. Nonetheless, spherical-shaped AgNPs were generated by the plant extracts of Prunus africana and Camellia sinensis. Easy, fast, high, and stable synthesis of AgNPs utilising plant extracts was also significantly impacted by a number of other parameters, including the extract salt ratio, incubation period, temperature, pH, etc. Chemistry involving reduction and oxidation is likely to play a role in the synthesis of AgNPs by plants. Vitamins, amino acids, proteins, enzymes, organic acids, flavonoids, terpenoids, alkaloids, polyphenols, and polysaccharides are all believed to be present in the plant extract. These compounds play important roles in reducing silver salts and also act as capping and stabilising agents.

Production of AgNPs via Bacterial Mediation

Toxic and costly chemicals and the high energy needs of physiochemical methods can be avoided by using microorganisms as biological agents for the easy, cost-effective, and environmentally friendly synthesis of AgNPs. Because of their quick growth, simple cultivation, and convenience of handling, various microorganisms like fungi, algae, bacteria, and yeast are frequently chosen for the green synthesis of AgNP. The extracellular and intracellular approaches are two ways that microorganisms can be used for the environmentally friendly synthesis of AgNP. There are a wide variety of primary and secondary metabolites that microbes produce, both inside and outside of their cells. These include amino acids, enzymes, proteins, sugar molecules, organic compounds, and many more. We still don't know the whole process of how microbes biosynthesize AgNP. Much like plant-mediated synthesis, the chemistry of reduction and oxidation is the generally acknowledged mechanism of microbe-mediated production of AgNPs. In the first step, enzymes produced by microbes, such as reductase, lower the metal ions to NPs. Afterwards, a variety of biomolecules found both inside and outside of cells of microbes act as glues and caps [23]. Upon contact with the bacterial culture supernatant at 30 ◦C, nanoparticles were produced within 48 hours when 1 mM AgNO3 interacted with it. According to the results of the FE-TEM study, the synthesised AgNPs have a size ranging from 15 to 55 nm. Within 48 hours of the reaction, spherical-shaped nanoparticles ranging in size from 10 to 40 nanometers were produced by extracellular production of AgNPs utilising the culture supernatant of a bacterial strain Cedecea sp. The supernatants of Citrobacter spp. cultures were used for the quick production of AgNPs (in 180 minutes). Version 5. Another study shown that the culture supernatant of Penicillium chrysogenum bioreduced AgNO3 to produce AgNPs. The capacity to synthesise AgNPs with enhanced antibacterial activity against harmful pathogens has been demonstrated by Sphingobium sp. MAH-11 and Pseudoduganella eburnea MAHUQ-39. Rapid intracellular AgNP production by use of Streptomyces strains (in a matter of 5 minutes). Methods for the intracellular production of AgNPs by use of Cyanobacteria Desertifilum. Nanoparticles with a spherical form and a size between 4.5 and 26 nanometers were discovered. Easy, quick, and environmentally friendly synthesis of AgNPs was also achieved by using a variety of algae and fungus. For example, AgNPs ranging in size from 60 to 100 nm were synthesised using the culture supernatant of Aspergillus terreus.

Characterization of Green Synthesized AgNPs

Analysing the size, shape, purity, surface chemistry, morphology, and other properties of AgNPs is a crucial part of green synthesis. For the purpose of characterising green synthesised AgNPs, a number of instruments have been employed, including UV-visible spectrophotometry, X-ray diffraction (XRD), scanning electron microscopy, transmission electron microscopy, Fourier transform infrared spectroscopy, dynamic light scattering, zeta potential analyser, and others. At first glance, the colour shift indicates that AgNPs have been synthesised. Synthesis of AgNPs is typically indicated by a reaction mixture that is dark brown in colour. The next step is to use UV-visible spectrophotometry to determine that AgNPs have indeed formed. A prominent peak at around 400-470 nm was observed in the UV-visible spectrophotometry of the synthesised AgNPs. The size, shape, and morphology of the biosynthesized AgNPs determined the absorption spectra. The nanoparticles can be characterised with the help of strong equipment like SEM and TEM. To examine the size, shape, morphology, level of particle aggregation, and purity of produced nanoparticles, SEM and TEM are employed. The crystallinity, size, and other structural properties of nanoparticles can be studied using XRD, an analytical technique. We examine the hydrodynamic size and polydispersity index of the synthesised nanoparticles using dynamic light scattering (DLS). To ensure the stability of AgNPs in water-based suspensions, Zeta potential measurement is crucial. In general, AgNPs exhibit good stability when their Zeta potential is either more than +25 mV or less than -25 mV. FTIR spectroscopy is a powerful method for studying the proteins that stabilise and cap nanoparticles. Akter and Huq reported the biosynthesis of AgNPs utilising the culture supernatant of Sphingobium sp. MAH-11. They characterised the nanoparticles using UV-vis, TEM, XRD, DLS, and FTIR. A change in colour to a dark brown was the first sign of AgNP synthesis; a strong peak at 423 nm in the UV-vis portion of the spectrum verified the synthesis. The size ranged from 7-22 nm, and the TEM investigation confirmed that it was spherical in shape. The crystalline character of the synthesised AgNPs was evidenced by the crisp rings that were exhibited in the SAED pattern. The XRD pattern further demonstrated that the AgNPs were crystalline. Several biomolecules served as reducing agents, capping agents, and stabilising agents during the synthesis process, as revealed by the FTIR spectra.

Use of Environmentally Friendly AgNPs for Antimicrobial Purposes

The current surge in interest in nanoparticles is largely attributable to their potential to significantly enhance the efficacy of treatments for bacterial infections and illnesses. Silver nanoparticles (AgNPs) are one type of applied nanoparticle that has demonstrated promising results in combating infectious diseases and other microorganisms. Researchers are once again looking at the therapeutic capabilities of AgNPs systems as possible antibacterial agents [26] as a result of the fast rise of antibiotic resistance during this time. Articles in the scientific literature have suggested that silver nanoparticles (AgNPs) have antibacterial properties and have investigated this possibility as a potential method for creating silver-coated medical devices, silver-based dressings (nanogels, nanolotions, etc.), and antimicrobial control systems. Because of their larger surface area and lower particle size, AgNPs have a large contact area with the microorganisms. The biological and chemical capabilities of AgNPs are significantly enhanced by these traits, which in turn makes them a powerful material for killing bacteria [145]. Research also demonstrated that AgNPs stifled the expansion of E. coli ATCC-15224 in both liquid and solid cultures. The E. coli bacterial strain was completely killed by AgNPs at a concentration of 60 µg/mL, and even at that lower dosage, the growth and multiplication of bacterial cells were hindered. In recent times, AgNPs have been created by combining bacteria with diverse plant components; these AgNPs have subsequently been tested against a range of harmful pathogens, including some that are resistant to multiple drugs. Synthesis of AgNPs by bacteria and their antibacterial action against harmful Klebsiella pneumoniae and Salmonella enteritidis strains. In contrast to several commercial antibiotics like erythromycin, penicillin, vancomycin,

oleandomycin, novobiocin, and lincomycin, the synthesised AgNPs exhibited a 17.6 mm ZOI against K. pneumoniae and a 16.8 mm ZOI against S. Enteritidis, respectively. The synthesised AgNPs had MIC values of 12.5/50.0 µg/mL against K. pneumoniae and MBC values of 25.0/50.0 µg/mL against S. Enteritidis, respectively. When tested against Klebsiella pneumoniae and Salmonella enteritidis, these MIC/MBC values were significantly lower than those of other antibacterial agents, such as zinc oxide and gold nanoparticles. Their antibacterial effectiveness against pathogenic strains of S. typhimurium and Y. enterocolitica was studied in another investigation that demonstrated Arthrobacter bangladeshi mediated green production of AgNPs. With respect to Y. enterocolitica and S. typhimurium, the green-synthesised AgNPs exhibited a 20.4 mm and 18.3 mm ZOI, respectively. The synthesised AgNPs had MIC/MBC values of 6.2/12.5 µg/mL against S. typhimurium and 3.1/12.5 µg/mL against Y. enterocolitica, respectively. In comparison to other antimicrobial drugs, these MIC/MBC values against S. typhimurium and Y. enterocolitica were much lower. A study was conducted to assess the antibacterial efficacy of AgNPs synthesised using dried leaf extract of Tasmanian flax-lily. The microorganisms tested included S. aureus, S. epidermidis, P. aeruginosa, and Candida albicans. The green synthesis of AgNPs was carried out using extract from Chlorophytum borivilianum callus, and the resulting nanoparticle was tested for its antibacterial effectiveness against human pathogens including E. coli, S. aureus, B. subtilis, and P. aeruginosa. This finding demonstrated that the produced AgNPs exhibited potent inhibitory action against the examined microorganisms. In addition to P. aeruginosa, S. aureus, E. coli, and K. pneumoniae, it has been found that Prunus africana, Camellia sinensis, and Plantago major may all produce small-sized AgNPs. Research has shown that NPs with a smaller size exhibit stronger antibacterial activity, perhaps as a result of their increased surface area. Remarkably effective against antibiotic-resistant strains of S. aureus, E. coli, and P. aeruginosa were reported to be AgNPs. We synthesised AgNPs using an extract from purple heart plant and tested their antibacterial efficacy against Staphylococcus aureus and Escherichia coli. In comparison to the purple heart plant leaves extract, the synthesised AgNPs mediated by the extract exhibited much stronger antibacterial action against E. coli and S. aureus. The possibility of using biosynthesized AgNPs as an antibacterial agent against MDR bacteria was also suggested in another study, which found that they had outstanding antimicrobial activity against a variety of Gram-negative and Gram-positive pathogenic microorganisms [27]. A number of recent investigations have shown that bactericidal drugs can be conjugated with AgNPs to increase their bactericidal efficacy while decreasing their toxicity to mammalian cells. Antibacterial drugs' therapeutic efficacy against bacterial infections can be improved through conjugation, which increases the amount of the agent at the specific bacterial location. Clinical trials with AgNPs against human pathogenic strains of Candida albicans, Salmonella enterica, Escherichia coli, and Vibrio parahemolyticus have also been described. The antibacterial effectiveness of AgNPs produced using green synthesis against pathogenic microorganisms that are resistant to many drugs was proven. As an antimicrobial agent to manage drug-resistant bacteria, it may be used in both medical and non-medical sectors. It was also claimed that it was inexpensive, eco-friendly, safe, facile, and effective. When tested against MDR-pathogens including S. aureus and P. aeruginosa, the biosynthesized AgNPs showed impressive antibacterial activity. S. aureus and P. aeruginosa were inhibited by MBCs at concentrations of 200 and 50 µg/mL, respectively. While certain commercial antibiotics like penicillin G, erythromycin, oleandomycin, lincomycin, and vancomycin either did not work at all or had very weak antibacterial activity against V. parahaemolyticus and S. Typhimurium, respectively, the author of this other study suggested that AgNPs produced by strain MAHUQ-40 exhibited substantial antibacterial activity. Antimicrobial efficacy against P. aeruginosa and Gram-positive B. cereus was the subject of another research. The pathogenic strains B. cereus and P. aeruginosa were inhibited in their growth by the synthesised AgNPs mediated by bacteria, which formed a distinct zone of inhibition. This killing power is what makes AgNPs so effective against germs. In addition, the surface modification of AgNPs through chemical functionalization has garnered significant attention and has the potential to be applied in several fields, including medicine, engineering, and biology, and it has improved the interactions between the constituents.

Mechanisms by Which AgNPs Fight Bacteria

When it comes to nanoparticles, the most crucial factor is their action mechanism, which is highly sensitive to factors like medium size, pH, and ionic strength, as well as the capping agent type. To this day, however, the exact antimicrobial action of AgNPs remains a mystery. Based on the findings of earlier research, one possible mechanism for the bactericidal action of AgNPs could be their frequent release of silver ions (Ag+). Evidence suggests that the antibacterial effects of the Ag+ ion are due to its ability to form complexes with nucleic acids and interact with their nucleosides. Sugars, proteins, and nuclear material were released through the ruptured membrane, demonstrating that nanoparticles changed the permeability of the membrane. Ag+ ions were able to cling more firmly to bacterial cytoplasm and cell membranes due to electrostatic attractions and an affinity for sulphur proteins, which in turn caused bacterial cell wall breakdowns and increased permeability. When cells release free Ag+ ions, it can disrupt the release of adenosine triphosphate (ATP) and lead to an increase in reactive oxygen species (ROS). These reactive oxygen species (ROS) have the potential to induce DNA-related problems, such as DNA replication and cell proliferation, by rupturing cellular membranes and altering deoxyribonucleic acid. In contrast, denaturing cytoplasmic ribosomal components may be an effective way for free Ag+ ions to disrupt protein synthesis. Because of their nanoscale size, AgNPs can penetrate bacterial cell walls and denaturate their cell membranes, hence inhibiting bacterial growth. This effect is due to the release proportion of Ag+ ions. Denaturation of the bacterial cell membrane can lead to cell lysis by rupturing the membrane and releasing intracellular and extracellular components. There is a strong correlation between the synthesis and processing parameters, as well as the dissolution profile of the AgNPs in the reaction media, and their antibacterial mechanisms .

The study's authors postulated that biosynthesized AgNPs would alter bacterial cell shape, infiltrate cell membranes, and cause cell wall damage in E. coli and Staphylococcus aureus, leading to reduced cell reproduction and eventual cell death. The FE-SEM images demonstrated that AgNPs have a strong antibacterial action and can effectively combat harmful bacteria. This has led to the increased use of AgNPs as an antimicrobial agent [28]. Synthesised AgNPs altered the structural function of

bacterial cells, according to another study. This included S. aureus and P. aeruginosa. Because of their mechanical properties, the suggested AgNPs have great potential as an antibacterial agent with a wide range of therapeutic uses in the fight against Pseudomonas aeruginosa and Staphylococcus aureus infections. It was shown that AgNPs had the ability to change the shape of cells and destroy their membranes in the pathogens that were tested. The primary action of AgNPs is to promote DNA damage, which causes the nanoparticles to firmly adhere to the surface of bacterial cell membranes and disrupt their normal function. The capacity to cross the cell membrane is another property of AgNPs. Once inside, they can react with complexes mediated by phosphorus, such as deoxyribonucleic acid, and sulphur, which can disrupt biological components. Research using scanning electron microscopy (SEM) and transmission electron microscopy (TEM) showed that AgNPs may bind to E. coli, interact with the bacteria, and enter their cells. The overall surface area of the nanoparticles is responsible for their antibacterial activity, which is enhanced by their adhesion and interaction ability. Consequently, there are a number of potential pharmaceutical and non-pharmaceutical uses for AgNPs synthesised in an environmentally benign manner, including the elimination of drug-resistant microbes.

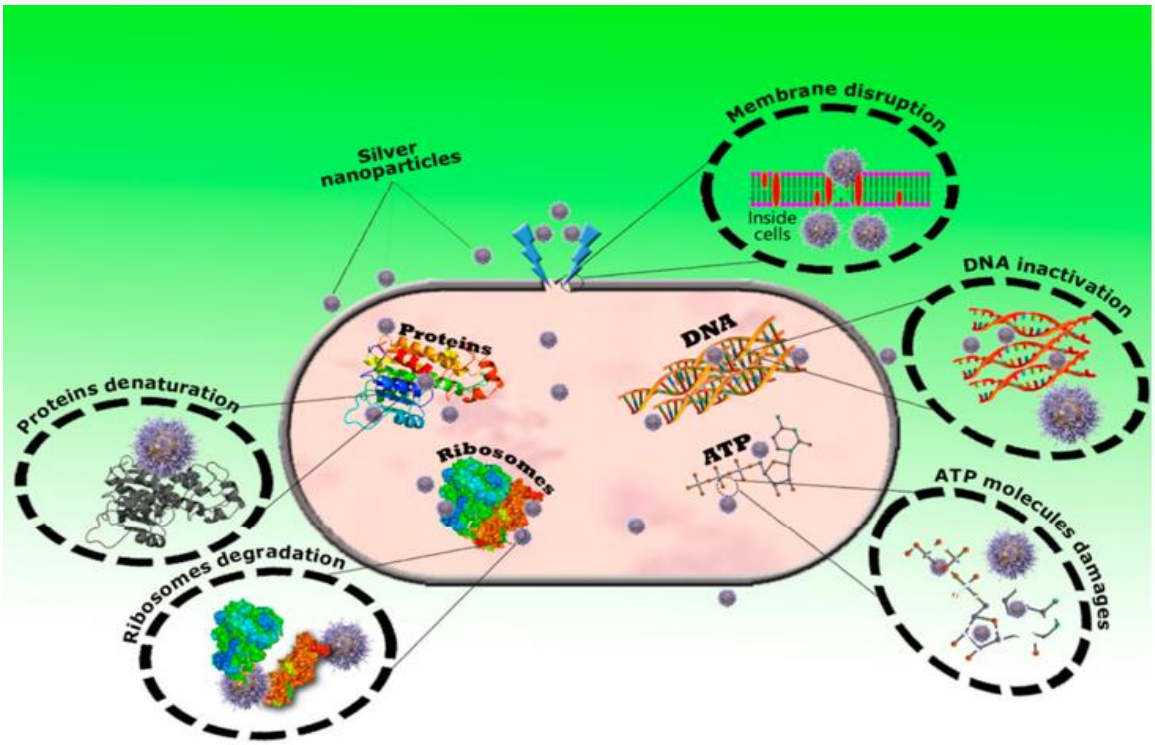

Figure 4: Exploring the potential antimicrobial actions of AgNPs. Damage to ATP molecules caused by reactive oxygen species generation, DNA inactivation, protein denaturation, ribosome breakdown, cell wall and membrane disruption

Advancements in nano biotechnology: synthesising algal nanoparticles

Like other biological organisms such as mushrooms, yeast, and bacteria, the study of algae for nanoparticle synthesis has expanded into the field of phyco-nanotechnology. The utilisation of microalgae in nanoparticle production has been underexplored, in contrast to the many research that have concentrated on seaweed extracts. Both macro- and microalgae are at the forefront of the race to find versatile uses for metal nanoparticles, according to recent investigations. In addition to their use in nanoparticle production, algae have several agricultural uses as fertilisers, particularly seaweeds, which reduce nitrogen and phosphorus discharge more effectively than conventional animal dung. Because of their malleability and exceptional capacity to hyperaccumulate heavy metal ions, algae are being considered as potential model organisms for bio-nanomaterial processing. There is still unrealised potential in less-studied algae species, even though progress has been made in synthesising algal nanoparticles. Clarifying the proteins and enzymes required to produce algal nanoparticles and easing the complexities of these reactions should be a goal of future research efforts. The development of commercially viable methods for nanoparticle synthesis should be a primary goal, hence the emphasis should shift to their design. Comprehensive investigations into the biological capacities of algae are necessary due to the revolutionary potential of nano-biotechnology in human health, agriculture, and the food business. To realise the whole range of uses, it is crucial to study their behaviours, collect data from plants and animals, and develop practical ways for synthesising nanoparticles [30, 31]. The chemical composition and size distribution of nanoparticles generated from algae need to be further investigated in future research. Nanoscale biotechnology that uses algae to synthesise nanomaterials is in its early phases and needs more work before it can be fully utilised.

CONCLUSIONS

Because it doesn't harm the environment, green synthesis of AgNPs is the way to go. One easy, efficient, and eco-friendly technique to synthesise AgNPs is to use different parts of plants, bacteria, fungi, and algae. Amino acids, proteins, enzymes, terpenes, alkaloids, flavonoids, phenols, tannins, vitamins, and many other biomolecules that serve as reducing,

capping, and stabilising agents are found in plant extracts. In a similar vein, microbes produce a wide variety of biomolecules both within and outside of their cells, including enzymes, amino acids, proteins, and a plethora of secondary and primary metabolites that serve as reducing agents, capping agents, and stabilising agents. There is promising evidence that biosynthesized AgNPs can kill a wide range of bacteria, including both Gram-positive and Gram-negative strains. Also discussed are the biosynthesized AgNPs' antimicrobial uses and how they work against harmful microorganisms. The precise process of synthesis and the manner of antimicrobial action remain poorly understood, despite the promising results of the fast, easy, and environmentally friendly synthetic approaches involving plants and bacteria in AgNPs. Therefore, there are a number of factors that could be taken into account when synthesising AgNPs from plants or microbes in the future. Choosing plants or microorganisms for synthesis that is simple, quick, and environmentally safe is the first step. Researchers need to think about the accessibility of plants and how to remove them while making plant selections. For the facile and mass manufacturing of AgNPs, plants should be readily available and the extraction method should be straightforward. Similarly, for the sake of safety and ease of handling during synthesis, researchers should concentrate on non-pathogenic, rapidly growing microorganisms. Here, probiotic bacteria may out to be the most effective synthetic ingredient. Next, we will look into the biomolecules found in plant extracts, microbial biomass, and culture supernatant. Many biomolecules found in plant extracts or microbial culture supernatant are thought to play a significant role in the production and stabilisation of AgNPs. There has to be extensive research on the function of different enzymes in biosynthesis. It is also the job of these biomolecules to make the synthetic AgNPs more effective against bacteria. In order to successfully synthesise AgNPs, it is crucial to study the biomolecules found in plant extract or microbial culture supernatant. Lastly, achieving fast, stable, and mass-produced AgNPs through parameter optimisation. A number of studies have shown that the synthesis process is highly sensitive to factors like incubation duration, temperature, pH of the reaction, concentration of plant extract and AgNO3, and so on. Because of this, optimising these reaction conditions allows for industrial-scale mass production. The study of antimicrobial mechanisms comes up at number four. Studies have shown that AgNPs are effective at the screening level, but few have looked into the underlying mechanisms. Understanding how AgNPs work against infections is crucial. Fifth, studying the human cell viability impact of biosynthesized AgNPs. According to certain research, AgNPs can kill human cells. So, to make sure biosynthesized AgNPs are safe for humans and the environment, it's important to study their possible toxicity to human cells.

A more environmentally friendly option to traditional chemical methods for producing nanoparticles is the use of plant extracts, microbes, and algae as synthesis platforms. Toxic reagents and harmful by-products can be reduced by the use of biologically mediated techniques that synthesise a variety of metal and metal oxide nanoparticles, such as silver, gold, platinum, palladium, copper, and iron. The higher biocompatibility of the resultant nanoparticles makes them appealing for use in biomedicine and pharmaceuticals. In addition, by optimising reaction conditions and meticulously selecting biological sources, biological systems provide controlled manipulation of nanoparticle size, shape, and surface properties. The unique ability of algae to accumulate metals and their abundant bioactive compounds make them ideal nano-factories, with uses ranging from agriculture and environmental remediation to medicine. Maintaining product uniformity, establishing structure-property connections, and ensuring reproducibility all need comprehensive characterisation employing techniques including TEM, SEM, XRD, FTIR, and UV-Vis spectroscopy. It is crucial to do toxicological profiling and thorough safety evaluations to confirm long-term biocompatibility and minimise unexpected biological risks in order to have successful clinical and industrial translation. Scalable production processes, regulatory compliance, and batch-to-batch standardisation are also critical for commercialisation and worldwide adoption. Assuming future development frameworks incorporate repeatability, safety validation, and regulatory readiness, green synthesis offers a sustainable and adaptable method for next-generation nanomaterials.

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