

## Synthesis of Silver (Ag) nanoparticles and their antimicrobial activities

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### Abstract

The problem of antimicrobial resistance (AMR) has become one of the most important health issues worldwide, as it has placed the efficacy of traditional antibiotics under a very strict limitation, compelling us to seek alternative methods of combating the problem. Silver nanoparticles (AgNPs) have found considerable application among other nanomaterials, as they exhibit broad-spectrum antimicrobial effects, remarkable physicochemical behaviour and possibly overcome drug-resistant pathogens. The present review gives a detailed discussion on silver nanoparticles with specific focus on the methods of their manufacture, action mechanisms and the biomedical applications of the nanoparticles. Traditional chemical, physical and photochemical processes of AgNP synthesis are also mentioned, as well as environmentally-friendly biological methods, such as microorganism- and plant-mediated green synthesis. The antimicrobial activity of the AgNPs against bacteria, fungi, viruses, and biofilms is critically evaluated with special emphasis on the critical parameters; the particle size, shape, concentration, surface charge, and pH in the environment. There is also the discussion of the synergistic outcomes of the AgNP-antibiotic conjugates that allow to have lower doses of antibiotics and establish the activity of the traditional drugs against the resistant strains. In addition, the mechanisms of antimicrobial action, such as disruption of membrane, the formation of reactive oxygen species, and release of silver ions are studied in detail. Lastly, the existing and future uses of AgNPs in biomedicine, consumer products, textiles, and in environmental remediation are presented, as well as biosafety and toxicity concerns.

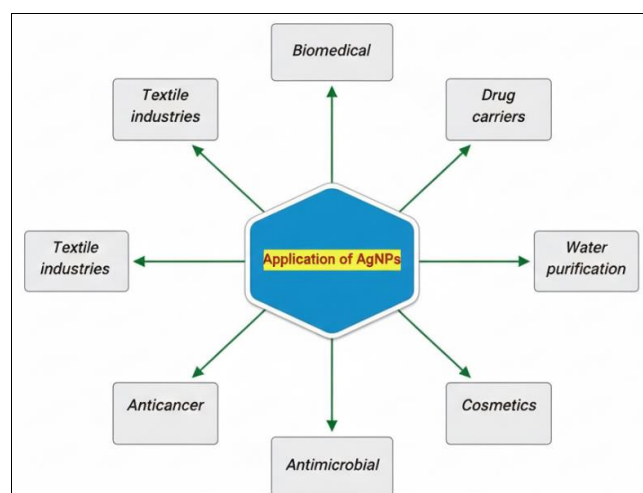
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### Introduction

Antimicrobial resistance (AMR) is an increasingly important health issue globally, which impacts greatly on the population health, agriculture, and economy everywhere. Antimicrobial drugs widespread use in agriculture, livestock production, and healthcare has resulted in the appearance of germs that are resistant. AMR is a complicated and pressing global health issue that calls for a diversified strategy. The delicate balance that has characterised modern medicine over the past century between microbes and antibiotic medicines is upset by AMR (Aijaz *et al.*, 2023) [3].

Silver compounds were frequently used to treat infections and prevent infections during surgery prior to the commercial manufacturing of antibiotics. Chemically and biologically manufactured nanosilver has been used since the 20th century in both medical treatments and commercially available, FDA-approved or non-approved goods such cosmetics, facemasks, and apparel. However, more research is required to fully understand the antibacterial mechanism of nanosilver and how it depends on its physicochemical characteristics. By concentrating on the antibacterial advantages of silver nanoparticles without (AgNPs) and with antibiotic functionalisation (AgNP-antibiotic conjugates) against pertinent Gram-positive and Gram-negative bacteria, this review sought to close this gap. After more than a century of producing nanosilver, methods for creating AgNPs utilising chemical reducing agents and extracts from bacteria, fungi, or plants are well known. AgNPs antibacterial properties and the ensuing antimicrobial synergy are also well-established. It is clear that synergy enables the use of lower antibiotic concentrations, which may lessen the rise in the emergence of microbes resistant to antibiotics. Additionally, as the

silver nanoparticle drug complexes can more readily elude microbial defences, older medications may be reused by conjugating them to AgNPs (Kaiser *et al.*, 2023) [18].



The scientific field that studies the manipulation of materials at the nanoscale is called nanotechnology. It broadly refers to the creation, modification, and characterisation of materials and structures with one or more dimensions at the nanoscale, or less than 100 nm (Santos *et al.*, 2015) [38]. Due to its applicability in many areas of human welfare, nanotechnology has advanced significantly over the past 20 years to become one of the most studied and rapidly expanding fields. Nanoparticles (NPs) are incredibly small particles, between 1 and 100 nm, that are either naturally occurring or artificially created. They have special and useful chemical and physical characteristics. Particles exhibit improved catalytic, magnetic, electrical,

mechanical, optical, chemical, and biological properties at the nanoscale. NPs exhibit greater reactivity, mobility, dissolving characteristics, and strength as a result of their high surface to volume ratio (Sahoo *et al.*, 2021) [36]. Iron, gold, silver, titanium, cerium, platinum, thallium, and other metals are among the several kinds of metal nanoparticles (Piñón-Segundo *et al.*, 2013) [33]. Because of their unique physicochemical and biological characteristics such as their high surface area to volume ratio, superior surface Plasmon resonance, ease of functionalisation or conjugation with various ligands to obtain desired tailored properties, toxicity against pathogens, effective cytotoxicity towards cancer cells, catalytic applications, and more silver nanoparticles are one of the metal nanoparticles that have been thoroughly investigated for a variety of scientific purposes. (Akter *et al.*, 2018) [4].

When compared to other similar procedures, green manufacturing of nanoparticles employing living cells through biological pathways is more efficient and produces a larger mass. Numerous components and biochemicals that can function as stabilising and reducing agents to synthesise green nanoparticles can be found in plants. In comparison to other biological, physical, and chemical approaches, the green synthesised technologies are more stable, economical, non-toxic, and environmentally benign. The three categories of green synthesis of nanoparticles are phytochemicals, intracellular, and extracellular. Because plant extract contains a large number of phytochemical components that can also function as reducing and stabilising agents, turning metal ions into metal nanoparticles, the synthesis of nanoparticles from plant extract is an economical approach that yields greater yields. Emerging uses for green synthesised metal and metal oxide nanoparticles in the biomedical field include bio sensing, wound healing, tissue treatment, immunotherapy, regenerative medicine, and dentistry (Vijayaram, *et al.*, 2024) [43].

### Synthesis Methods of Nanoparticles

There are many approaches used to synthesize nanoparticles-

#### a. Chemical Method

The most practical, efficient, and manageable way of creating nanoparticles is chemical synthesis. The most popular method for creating nanoparticles is the chemical reduction process, which uses both organic and inorganic reducing agents. Silver ions ( $\text{Ag}^+$ ) in both aqueous and non-aqueous solutions are generally reduced using a variety of reducing agents, such as ascorbate, polyol process, sodium borohydride ( $\text{NaBH}_4$ ), sodium citrate, poly (ethylene glycol)-block copolymers, N-dimethylformamide (DMF), Tollens reagent, N, and elemental hydrogen. Such substances react with the silver nanoparticles, reducing  $\text{Ag}^+$  and the results are metallic silver ( $\text{Ag}^0$ ). The particles then group together to form oligomeric clusters, which eventually results in the creation of silver particles. Three processes make up the chemical methods for silver nanoparticles in real solution: Precursors based on metals, capping oxidants, and reducing agents. It is crucial to use capping agents to assist regulate the growth of nanoparticles during the synthesis process in order to produce metal-based nanoparticles that are uniformly spherical in shape and smaller in size. Two phases of growth and nucleation are involved in the creation of a colloidal solution of silver nanoparticles, which dictate the final nanoparticles' size and

shape. However, the regular growth rate and size in the production process is critical to the synthesis of the right size and homogenized nanoparticles. The production of growth rate and size similarity are key factors in the synthesis of single dispersed nanoparticles of regular size (Mughalet al., 2022) [26].

#### b. Physical Method

The process of condensation or vaporisation is used to create nanoparticles by keeping the furnace tube at atmospheric pressure. There are two methods for physically synthesising AgNPs: evaporation-condensation and laser ablation.

The production of nanomaterials in a furnace has several disadvantages, including cylinder-shaped particles occupying a large area and high energy consumption that significantly raises the ambient temperature. Furthermore, achieving thermal stability takes a long time. The unique cylinder-based furnace requires a heating period of 10 minutes in order to achieve a constant temperature because it requires many kilowatts of energy. Moreover, another way to create AgNPs is via laser ablation of bulk materials in a solution. The laser-based method is better than the other method since it produces pure colloids without the need for a chemical reagent (Hwang *et al.*, 2022). This kind of process creates a large quantity of product in a single step and produces more silver nanoparticles in the form of ash. AgNPs that do not originate at the base metal source are the only affected by a pulse laser when concentrated on a liquid system; no involving ions, compounds, or reducing agents. The type of base metal supply, length of irradiation, laser power, and liquid media properties are some of the factors that affect the properties of synthesised metal nanoparticles. Because laser ablation uses mild surfactants in the solvent rather than chemical reagents, it produces NPs in pure form without contamination (Chugh *et al.*, 2018) [9].

#### c. Photochemical Method

This is done by subjecting the metal precursor solution to visible or ultraviolet (UV) light. Compared to other procedures, photochemical processes of creating metallic nanoparticles are highly beneficial since they do not involve the use of toxic or harmful materials, they do not demand the use of costly equipment or highly trained staff, and they can be performed in an ordinary ambient setting, i.e. room temperature and atmospheric pressure (Santos *et al.*, 2015) [38]. Photochemistry is used to initiate the reduction of a metal precursor by adjusting the reducing agent (which is in an  $n + \text{valence state (Mn}^+)$  to a zero-valence state ( $\text{Mn}^0$ ). In order to make metallic NPs, the  $\text{M}^0$  produces nuclei or nucleation centres that grow and accumulate. Stabilisers and capping agents are both required to avoid agglomeration, enhance colloidal stability as well as control the production of homogenous metallic NPs in terms of desired shape and size (Zare-Bidaki *et al.*, 2022) [44].

#### d. Biological method

Metal nanoparticles synthesis biologically has gained a lot of interest in the recent past due to its diverse applications. It employs biological systems, like microorganisms (fungi or bacteria), and extracts of plants are perceived to be valuable alternatives to alternative synthesis systems.

### a. Microorganisms were used to mediate silver NPs synthesis

Microorganisms like bacteria and fungi remediate the toxic pollutants by means of the biological synthesis of metal nanoparticles and reduction of metal ions. Some bacteria can make AgNPs intracellularly in which case intracellular components can be used as stabilisers and reducers. The most complex physio chemical process can be replaced by an attractive approach incorporating the use of naturally available reducing agents as a way of physiologically synthesising the AgNPs. Green synthesis involves the employment of natural capping agents to stabilise AgNPs but is better than the methods before since it does not involve the use of dangerous chemicals and by-products (Patra *et al.*, 2015) [31].

### b. Silver NPs are produced by plants

**Plant-mediated synthesis of NPs can occur in 3 different ways:** (i) Intracellularly (within the plant), (ii) Extracellularly (with plant extracts), and (iii) with individual phytochemicals. There are many plants which can store the metals and then transform them within the cell to nanoparticles. Some of the biomolecules present in plants which are vital in reducing metals include amino acids, alkaloids, aldehydes, flavones, ketones, proteins, phenolics, polysaccharides, saponins, tannins, terpenoids and vitamins (Nath and Banerjee 2013) [28]. The variations in the stabilising and reducing potential of the biomolecules present in the plant lead to a variation in the size, shape and nature of accumulated NPs. (Gardea-Torresdey *et al.*, 2002) [15]. The plant extracts are prepared using several methods with some of them being hot extraction, cold extraction and use of Soxhlet apparatus which was later used in the production of NPs. This method of synthesising NPs is more suitable than the intracellular technique, since it is simpler to scale up and downstream. It is also environmentally friendly; green, biocompatible, renewable, and non-toxic. It has been identified that these NPs have numerous biological applications as they are biocompatible. To initiate the process of making metal nanoparticles the plant extract is introduced into the metal precursor solution that contains the respective metal salts. Metal precursor solutions AgNO<sub>3</sub> are usually utilized in creating Ag NPs. The first process of making metal nanoparticles using plant extract involves three fundamental steps. The first stage is a reduction of metal ions (M<sup>+</sup> or M<sup>2+</sup>) to metal atoms (M<sup>0</sup>), and the subsequent step is the nucleation of the reduced metal atoms. At the second stage, though, thermodynamic stability rises along with the process of uniting small nearby NPs into larger particle sizes. At the final stage, the process is finalized and the NPs are provided with the final form (Makarov *et al.*, 2014) [16]. Presence of various active biomolecules in the plant extract plays a major role in reduction and stabilisation of metal ions in the solution (Ranthappa *et al.*, 2002). It is however difficult to identify the exact reducing and stabilising agents of NPs formation due to the presence of numerous phytochemicals in the plant extract (Si *et al.*, 2007) [40]. To synthesize AgNPs, Salih *et al.*, (2020) [37] used the plant extract of three different plants, including *Eucalyptus globulus*, *Ziziphus spina-christi*, and *Piper nigrum*. The mean distribution of particle size decreased with the increase in the content of plant extract and was in the range of 8-35 nm.

### Applications

#### Silver Nanoparticles have anti- microbial capability.

Health related issues have risen due to the resistance of the microbes to antimicrobials agents, including antibiotics. It has been shown that novel applications of metal nanoparticles can be discovered by combining the three forces of the material science, nanotechnology, and at the same time, the inherent antibacterial properties that certain metals possess (Allahverdiyev *et al.*, 2011) [5]. Many studies have been conducted to enhance the existing antimicrobial therapies given that the development of multi-drug resistance microbes is presenting a serious health risk to the population (Pelgrift and Friedman 2013) [32]. Approximately 7 out of 10 bacterial infections have been reported to be resistant to either one of the first and second line medications that used to be used in the past to treat the infection (Allahverdiyev *et al.*, 2011) [5].

#### Antibacterial activity

The use of silver as an antibacterial is not a novel phenomenon and its Nano forms are better and more biocompatible. Within the context of this field, much research has been conducted since the early days of the use of silver in nanotechnology. Many *in vivo* and *in vitro* studies have been carried out so far with regard to the antimicrobial potential of nanosilver or nano conjugates of silver. Such studies can assist us in knowing the importance and possibly the role of the silver nanoparticles in future and also in treating the microbial diseases by taking the micro molecular shortfalls of the classical antimicrobial preparations, combating drug-resistant pathogens, and also in laying the combinatorial approach basis of the action of drugs. The nanoparticle due to the additive effect of the nanosilver and a large selection of phyto constituents with inherent antimicrobial effect, silver nanoparticles can be easily prepared out of the plant extracts with excellent stability and environmental affordability and exhibit a broad spectrum of antimicrobial effect. A study by Loo *et al.*, (2018) [21] has shown that silver nanoparticles (size: 4.06 nm) prepared using pu-erh tea leaves possess strong antimicrobial action against gram-ve foodborne pathogens: *Salmonella typhimurium*, *Escherichia coli*, *Klebsiella pneumoniae*, and *Salmonella enteritidis*. also The reported MICs were 3.9, 3.9, 7.8, and 3.9 µg/ml.

This has been due to the long production-consumption cycle which has led to the bacteria developing resistance to the chemical antibacterial agents that are normally taken, bringing down the effectiveness. Moreover, the problem of antimicrobial resistance has enhanced drastically as the use of low-quality or off-the-shelf drugs has grown in the underdeveloped countries (Tenover & Hughes 1996) [42]. There is an urgent need to hasten the process of creating new and effective antibacterial agents and their production. The use of NPs as the antibacterial agents has proved to be a solution to this dilemma since they can create an effective nanostructure that can be used to deliver the antibacterial chemicals, therefore better and locally to address bacterial growth. Moreover, the same has shown that nanoparticles are so strong that the pathogens have few ways of getting resistance to them. The advantage of implementing metal oxide nanoparticles in a larger scale is that most of them do not pose any harm to mammalian cells at the dosages that have been used to kill bacterial cells (Dizaj *et al.*, 2014) [11].

Nanoparticles can also be characterized by the significant impact that they have on their antibacterial properties depending on their form. Such shape-related characteristics of nanoparticles were the focus of many studies. Antibacterial action was found to be present in Ag NPs of three distinct shapes of spherical, rod-shaped and truncated triangles. It was demonstrated that the triangular NPs were truncated and thus could be more active in antimicrobial activity due to the high density of atoms on its surfaces and thus this made them more likely to be reactive (Pal *et al.*, 2007) [30]. The researchers opined that such AgNPs can be used in various other procedures, such as covering some biomedical products, biofilms, wound dressing, and bio-adhesives (Allahverdiyev *et al.*, 2011) [5]. The exact mechanisms through which Nano metals are able to show their antibacterial activity are under research. Two standard options have been put out in this respect. To begin with, hazardous effects associated with free metal ions could occur when the metals have dissolved off the surface of NPs. Second, the surface of the NPs can generate reactive oxygen species (ROS) which has the potential of leading to oxidative stress. A study of the research suggests that nanomaterials have several inherent features that can influence the efficacy of nanomaterials in reducing or eliminating cells (Besinis *et al.*, 2014) [6].

#### Antifungal Activity

Even though biological AgNPs possess several benefits as compared to AgNPs synthesized through chemical and physical methods, AgNPs has been proposed as a potential solution. The growing conditions of the organism, the organism used in synthesis, the physicochemical properties of the AgNPs, as well as the target organism have an effect on the antifungal activity of the different biogenic nanoparticles. The extensive variety of methods used to evaluate the antifungal activity of these biogenic nanoparticles highlights the importance of applying the globally recognised methods to ensure that the techniques used have appropriate controls in order to obtain consistent and comparable results. In order to obtain meaningful data on the antifungal action of a new drug towards a specific species, it is important to have a large number of isolates of the same species since a heterogeneity of a species can occur as a result of genetic variation. The AgNPs action on fungi mechanism has been understood significantly. They have a great potential in being used as antifungal agents in clinical practice because they have shown that they can act on multiple targets. Biogenic AgNPs have potential applications as replacements to the toxicity of current drugs and the problematic disease caused by resistant fungus due to their broad spectrum of antibacterial effect and potential synergistic interactions with antifungal drugs. We believe that the biogenic AgNPs will be used as a general antifungal agent at low cost (Mussin & Giusiano, 2022) [27].

#### Antiviral Activity

The first important aspect is size with regard to the most studied type of nanoparticles, which are spherical ones. To be more precise we have shown that the antiviral effect is the most potent in the case of small size, which is usually about 10 nm, but the antiviral efficacy falls as the diameter grows. The second factor is concentration of the nanoparticles in the system; the greater the concentration of AgNPs the greater the effect. Because excess silver may

cause toxicity problems, size and concentration have a close connection. Presence or functionalisation of AgNPs in composite materials is the other factor that could influence the AgNP properties. In the latter case, the addition of intrinsically antiviral chemical agents/coatings may serve to emphasise the antiviral effect; otherwise, the presence of a matrix may also act as a physical shield against the interaction of the virus with the host cell. Moreover, some types of functionalisation can be able to mimic the receptors of cells, which improves the interaction of the functionalised AgNPs and viruses and prevents the attachment of viruses to cells. Finally, it is also possible that the duration of exposure can affect the antiviral activity of the system. Due to their virucidal action mechanism, which renders them the most common, AgNPs have a huge potential in the sphere of antiviral materials. The best applications of AgNPs are in the textile sector and water and air purification systems that may considerably enhance the quality of life of people. The development of antiviral devices to improve the efficacy of surgical masks or filter the air in particular may be a rational method to revert to normal life, especially at the current time when SARS-CoV-2 is spreading. These technologies could also be applicable in the future to fight the well-known viruses that are difficult to handle or those that could be transmitted like a wildfire (Luceri *et al.*, 2023) [22].

#### Anti-biofilm Properties

AgNPs appear to be the best anti-biofilm agents and most studied. The use of metal nanoparticles is one of the possible strategies that would help prevent the growth and formation of the biofilm by bacteria and fungi. In addition, metal nanoparticles also possess favourable antibiofilm qualities which lead to elimination of the developed biofilms. Further studies are required on the penetration of metal nanoparticles into different types of biofilms and whether they could have any long-term effects on the characteristics of biofilms, e.g. the production of metal ions in a slow manner over long periods of time. On the same note, more studies are needed to learn the mesoscopic interactions between metal nanoparticles and quorum-sensing. In a biofilm, metal nanoparticles have the ability to react with EPSs (Extracellular Polymeric Substances) and bacteria. Metal nanoparticles can be mixed with antibiotics or with another to increase their effects on the existing biofilms (Li *et al.*, 2024) [20].

#### Mechanism of AgNPs Antimicrobial action

The silver nanoparticles according to reports change the physiological and structural properties of cell membranes in the microbes such as build up, change in permeability and membrane potential and inhibition of respiratory proteins attached onto the membrane, disrupting the homeostasis of the cell and eventually causing cell death in the microbes. Also, it is suggested that antibacterial effects of Ag ions are linked to its positive charge (Abbaszadegan *et al.*, 2015) [1]. The antibacterial activity of AgNPs is largely affected by a number of factors, which include type of microbe, temperature, pH, and concentration of AgNO<sub>3</sub> (Marambio-Jones and Hoek, 2010) [24]. It has an inverse relationship with Ag + concentration. The reason is that the smaller particles will have more surface area to interact and hence they will be effective in killing bacteria compared with large particles that have a smaller surface area (Chanda, 2013) [8]. When the Ag + ions penetrate the bacterial cells and are

reduced to Ag<sub>2</sub>O, it is found that the toxicity to the bacterial cells is through the Ag<sup>+</sup> ions released by AgNPs (Hsueh *et al.*, 2015) [16]. Moreover, AgNPs can be released as internalised, producing Ag<sup>+</sup> ions, which bind to sulfhydryl groups of proteins, denaturing them. AgNPs are known to attach themselves to the respiratory chain of bacteria and disrupt crucial physiological properties that eventually lead to the death of the cell (Menu *et al.*, 2012) [25]. Most studies have found that silver nanoparticles are able to generate reactive oxygen species, including hydroxyl and superoxide radicals, resulting in oxidative stress to the cell and causing a severe damage or even death to the microbial cell (Rautela *et al.*, 2019) [34].

### Influencing Factors Antimicrobial Efficacy of AgNPs

The size, shape, concentration, and surface charges are the most important factors that determine the antibacterial properties of silver nanoparticles.

### Size, Shape and Concentration of AgNPs

The platelets, cubes, decahedrons, triangles, spheres, and numerous other forms have been found to exhibit the highest antibacterial activity; the triangular and the sphere shape is the most effective (Zorraquin-Peña *et al.*, 2020). AgNPs were obtained in large yield and monodispersity with means of 5, 7, 10, 15, 20, 30, 50, 63, 85 and 100 nm. Silver nanoparticles were identified to have minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) which were both dose and size dependent. Based on the results of the experiment, all the AgNPs are highly deleterious to various types of bacteria and their antibacterial activity becomes even faster as their particle size reduces to 5 nm, compared to 7 nm and 10 nm AgNPs at their respective minimum bactericidal concentration doses. The three bacterial strains that were used in the study are *S. aureus*, *E. coli* and *B. subtilis*. It is interesting to mention that despite these strains, 5, 7 and 10 nm AgNPs have shown the same level of bactericidal activity (Agnihotri *et al.*, 2014) [2].

### Surface charge of AgNPs

Neutral, positive, and negative surface charges AgNPs were achieved. The gram-positive (including *Streptococcus mutans*, *Streptococcus pyogenes* and *Staphylococcus aureus*) and gram-negative (including *Proteus vulgaris* and *Escherichia coli*) bacteria were tested on all synthesised Ag NPs to determine the antibacterial activity. Based on the results of the study, the antibacterial effect of positively charged AgNPs is greatest against gram positive and gram negative bacteria. Ag NPs with negative charge showed the minimal amount of antibacterial action, and the neutral Ag NPs neutral Ag NPs had an intermediate level (Abbaszadegan *et al.*, 2015) [1].

### Effects of pH

*Pseudomonas fluorescens* was left to be exposed to well-characterized AgNPs throughout a day in pH values of 6 to 9. The development of the control was not much influenced by the pH, but it developed slower at low PH. The controls used were only bacteria, silver nanoparticles, latex nanoparticles (negatively controlled); the nanoparticles are of a similar size but carry insignificant or no chemical toxicity, and no bacteria or silver nitrate (positive control). It is only when there is no SRHA (Silver River Humic Acid)

that silver nanoparticles will be toxic at a 2000 ppb concentration resulting in a reduction of the bacterial cell density by 90 per cent during the 24 hours of contact (Fabrega *et al.*, 2009) [14].

### Other applications

Besides the antibacterial applications of silver nanoparticles, several other applications ought to be mentioned. They can be used in such areas as biomedicine, consumer goods, agriculture, Anticancer, cosmetics, water and air purification, textiles, and cars (Kumar *et al.*, 2020) [19]. What makes silver nanoparticles useful as biomedical products is the fact that they are effective and powerful antibacterial agents. The extensive antibacterial spectrum of their applications, their long-term efficacy, and their durability together with their constant release of silver ions make them applicable in numerous consumer and biomedical applications. Antimicrobial textiles, which are produced in the textile industry under the use of silver nanoparticles. AgNPs are incorporated with textiles to provide them with some special characteristics such as colour performance or brightness enhancers, durability, sanitary, and disinfection (Deshmukh *et al.*, 2019) [10]. Silver nanoparticles are finding a wider application in consumer products of desirable and unique nature. Examples of personal health care products include deodorants, skin creams, body lotions, toothpaste, antimicrobial socks, kitchen scrubbers, antimicrobial toys, disinfecting sprays, pesticides, keyboard covers, air and water treatment/purification devices, detergents, face masks, car interiors, food packaging and many more (Shafique and Luo, 2019) [39].

Silver particles are of superior choice because of its possible heat conductivity, electrical conductivity, low cost in comparison with graphene or gold, and chemical stability to be used as adhesives and conductive inks compared to most other electrically conductive fillers (Ren *et al.*, 2015) [35].

### AgNPs Toxicity and Biosafety

AgNPs have been demonstrated to possess desirable properties including antibacterial and antifungal properties, but *in vitro* studies have equally demonstrated that exposure of such nanoparticles may have adverse and deleterious consequences on host cells or bacteria. In a toxicological experiment, the different cells and organs are exposed to different levels of the substances, and their response is observed within a set period of time. The proper dosage of the medication can be determined in combination with exposure limit in order to avoid adverse effects, median toxicity (MD50), and median lethal dose (LD50). The cytotoxic action of silver nanoparticles has mainly been explained in the context of oxidative stress and genotoxic outcomes.

Massive production of ROS leads to necrosis or apoptosis which culminates into cell death. A report that was conducted on alveolar macrophages showed that alveolar macrophages with silver nanoparticles of an average size of 15 nm had the most significant loss of mitochondrial functionality. AgNPs are toxic depending on size and shape (Carlson *et al.*, 2008) [7].

*In vitro* research has proven the harmful impacts of AgNP exposure to bacteria or cells. The primary mode of AgNP-induced cytotoxicity in *in vitro* studies is an increase in the production of reactive oxygen species (ROS). The size of the AgNPs, its concentration, and exposure duration are the

main factors that determine its cytotoxicity and genotoxicity. Exposure to silver nanoparticles leads to reduced levels of GSH (glutathione), lipid peroxidation, up-regulation of ROS-responsive genes, and subsequent rise in their proteins, which finally causes damage to DNA, apoptosis, and necrosis. An *in vivo* study indicated that exposure to silver nanoparticles has a number of implications on several crucial organs. In many non-mammalian models of animals, the exposure to silver nanoparticles leads to morphological defects, deformities, and reproductive issues (Du *et al.*, 2018) [13].

The most commonly utilized metal nanoparticles and they are present in most products, such as textile, cosmetics, antimicrobial dressings, and home water treatment systems. The biological use and safer production of nanosilver would increase due to the development of methods to synthesize silver nanoparticles by ecologically safe green methods (Noga *et al.*, 2023) [29].

### Conclusion

The growing cases of antimicrobial resistance have put pressure on the quest to find good alternatives to the traditional antimicrobial agents and silver nanoparticles have come in as one of the hopeful remedies of this situation. Due to their peculiar physicochemical characteristics, AgNPs display an antimicrobial effect on a wide range of bacteria, fungi, viruses, and biofilms, including those that exhibit resistance to the commonly used antibiotics. It is pointed out that in this review the antimicrobial behaviour of silver nanoparticle is directly correlated to the size, shape, surface charge, concentration of nanoparticles and the route of synthesis, which all affect the interaction of the nanoparticles with the microbial cells.

Synthesis methods that are eco-friendly, especially the use of plant extracts and microorganisms, have a high potential of being more advantageous over traditional chemical and physical synthesis methods. These environmentally-friendly production methods are inexpensive, reproducible and less toxic reagents are used thus enhancing the biocompatibility and sustainability of AgNPs. Moreover, silver nanoparticles conjugation with antibiotics is a significant improvement because it improves antimicrobial activity, allows reducing doses, and, possibly, prevents resistance development.

In spite of the mentioned benefits, one cannot ignore the issues of the possible toxicity and environmental effects of AgNPs in the long term. *In vitro* and *in vivo* research have shown that overexposure could cause oxidative stress, cellular damage, and toxicity on organs. A delicate balance between biosafety and antimicrobial effect therefore has to be established. Standardized synthesis protocol, detailed mechanistic investigation and extended toxicity investigation should be put at the forefront of future research to provide safe clinical and industrial use.

On the whole, silver nanoparticles have significant potential to be used as multifunctional antimicrobial agents. As more research is conducted on safety, reproducibility, and responsible use, the use of AgNP-based technologies will probably have a significant part to play in tackling the problem of antimicrobial resistance and developing modern antimicrobial strategies.

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