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A Review on Loaded Silver Nanoparticles to the Activity Treatment of Antimicrobial and Antibacterial Activity

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Abstract: *Silver nanoparticles (AgNPs) represent a promising advancement in nanotechnology, combining the unique properties of silver with additional functional agents such as drugs, polymers, or biomolecules. The review paper defines and describes their scope highlights their versatile applications across medicine, agriculture, environmental remediation, and industrial processes. Their ability to serve as antimicrobial agents, drug carriers, and diagnostic tools makes them a significant focus for modern research and innovative therapeutic strategies.The analysis shows that these nanoparticles can serve as effective drug delivery systems, antimicrobial agents, and diagnostic tools, confirming their relevance in modern nanotechnology.*

Keywords*:* Nanotechnology, Nanoparticles, Loaded Silver Nanoparticles

I. INTRODUCTION

NANOTECHNOLOGY

The prefix “nano” refers to one-billionth. When applied in the metric scale of linear measurements, a nanometer is one-billionth of a meter. The term “nano-technology is now commonly used to refer to the creation of new objects with nanoscale dimensions between 1.0 and 100.0 nm. The term also conceptually implies the ability to manipulate individual atoms or molecules as the building blocks of man-made nanoscale structures. The term “nanoscience” is used to refer to research at a nanoscale.[1]

The concept of nanotechnology was first introduced in 1959 when physicist Richard Feynman presented a presentation on making things at the atomic and molecular levels. Nanotechnology is now regarded as the most promising technology of the twenty-first century, and researchers have investigated it as a novel technique in medical research.[2]

Man has been looking for miraculous remedies to relieve sickness and injury pain for hundreds of years. Many academics feel that nanotechnology applications in medicine can be essential in achieving this.[3]

NANOPARTICLES:

Nano particles are efficient delivery systems for the delivery of both hydrophilic cand hydrophobic drugs. Nanoparticles are the submicron-sized particles, ranging 10–1000 mm. The major goal behind designing nanoparticle as a delivery arrangement is to control particle size, surface properties, and release of pharmacologically active agents in order to achieve the site-specific action of the drug at the therapeutically optimal rate and dose regimen.[4]

CLASSIFICATION OF NPs :-

Nanoparticles (NPs) are categorized into the following classes based on their shape, size, and chemical characteristics

- Carbon-based NPs
- Metal NPs
- Ceramics NPs
- Lipid-based NPs

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TYPES OF DIFFERENT METAL-BASED NPS:

Metal NPs are purely made of metal precursors. Due to well-known localized surface plasmon resonance (LSPR) characteristics, these NPs possess unique optoelectrical properties. NPs of the alkali and noble metals, i.e., Cu, Ag, and Au, have a broad absorption band in the visible zone of the solar electromagnetic spectrum. The facet, size, and shape-controlled synthesis of metal NPs are essential in present-day cutting-edge material.

- Silver nanoparticles (AgNPs)
- Copper nanoparticles (CuNPs)
- Gold nanoparticles (AuNPs)
- Aluminum nanoparticles (AlNPs)
- Iron nanoparticles (FeNPs)

SILVER NANOPARTICLES (AgNPs):

AgNPs are particles with a size range of 1–100 nanometers made of silver. They have unique physical and chemical properties due to their small size, high surface area-to-volume ratio, and ability to absorb and scatter light in the visible and near-infrared range. Because of their relatively small size and high surface-to-volume ratios, which cause chemical and physical differences in their properties compared to their bulk counterparts, silver nanoparticles may exhibit additional antimicrobial capabilities not exerted by ionic silver.[5]

LOADED SILVER NANOPARTICLES :-

Loaded silver nanoparticles are silver nanoparticles that have been functionalized, coated, or embedded with additional substances such as drugs, polymers, biomolecules, antibiotics, plant extracts, or other active agents.

They consist of:

- A silver nanoparticle core (providing antimicrobial, catalytic, or sensing properties), and
- A ‘loaded’ outer layer containing the functional material designed to enhance stability, targeting, or performance.

In simple terms:

Loaded AgNPs = silver nanoparticles + an added functional agent for enhanced or specialized application.

ANTIBACTERIAL ACTIVITY OF SILVER NANOPARTICLES

For the last 30 years, the pharmaceutical industries have focused on developing new antibiotics that have the ability to better fight against DNA replication, protein synthesis and bacterial cell wall synthesis. Despite this progress, a high fatality rate is still present due to the rise of antimicrobial resistance in bacterial infections. Bacterial resistance to the traditional antibiotics is a great healthcare issue with huge impact worldwide.

There is an increasing growth on using nano materials and they are becoming an important part of our lives. Efforts are made towards discovering non-toxic and cost-effective materials that can be used for various applications in industry, medical, pharmaceutical and cosmetic domains. An important application of such materials is drug-resistant bacteria and diseases control. Nanoparticles obtained from silver, gold, platinum and semiconductors can be used with success as delivery agents for carrying small molecules such as drugs. Nanoparticles became highly effective against bacteria due to their antimicrobial activity because of their large surface area allowing high synergy arising from multivalent interactions.

Among the variety of engineered nanoparticles that have been used in antibacterial treatments, silver (Ag) nanoparticles is the most widely used antibacterial nano agent because of its broad-spectrum antimicrobial properties and strong antimicrobial effectiveness against multiple bacteria, viruses, and fungi. The first evidence of using silver in medicine dates back to 1881, when it was used to prevent eye infections in neonates, and later on, in 1901 as internal antisepsis. Nowadays, drugs containing silver such as silver nitrate and silver sulfadiazine are commonly used to treat dermal burns, wounds and to remove warts.

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Scientists agree that Ag nanoparticles interact with the bacterial cell envelope, however it is still unknown what the primary cellular target is silver nanoparticles are antibacterial agents, capable of fighting against 650 types of diseases. Recently, many publications discovered proof for the antibacterial activity of Ag nanoparticles combined with common antibiotics, especially against multidrug resistant bacteria including *Staphylococcus aureus* and *Escherichia coli*.

An increased antibacterial activity can be observed when combining Ag nanoparticles and antibiotics, especially against drug-resistant bacteria. In recent times, this combination has been considered a potential method to overcome bacterial drug resistance. Binding to various antibacterial agents for obtaining a higher antimicrobial activity was proposed. Several studies have been studying Vancomycin capped with Ag nanoparticles. While some autors reveal improved effects against both Gram -positive (*S. aureus*) and Gram- negative (*E. coli*). Recent studies show that the nanomaterial is capable of developing a better antibacterial activity against Gram positive bacteria but not against Gram negative bacteria. During the First World War, silver was the most used substance in fighting and treating soldiers' infections.

Silver nanoparticles have proven a high antimicrobial activity and a low cytotoxicity level when compared to particles obtained from other heavy metals like gold, platinum and zinc. They can bind to cells and restrict enzyme activity, destabilize the cell membrane and eventually lead to cell death.

Studies have demonstrated that AgNP could induce: cyto-toxicity, geno-toxicity, inflammatory response, DNA damage, ultimately cell death. Cell's membrane and neuronal structures prolonged contact may lead to: skin diseases, argyria disease (blue skin)

Factors Impacting the Antibacterial Activity of Silver Nanoparticles :-

Antibacterial activity can be influenced by:

- Nanoparticle Size
- Nanoparticle Shape
- Nanoparticle Surface Chemistry
- Antibacterial Mechanisms Activated by Silver Nanoparticles[6]

Mechanism of Antibacterial Activity of Silver Nanoparticles :-

There are three mechanisms by which silver acts on microorganisms. One mechanism is that silver cations penetrate the cell wall of bacteria and react with peptidoglycans.

Oxidative stress, which results from the binding of AgNPs to a bacterial cell causing the ions' release, is the second form of antibacterial action of silver nanoparticles. Silver nanoparticles can bind to membrane proteins, which can significantly affect membrane permeability. This may result in leakage of cell contents, i.e., uncontrolled transport across the cytoplasmic membrane. AgNPs that bind to membrane proteins can affect the uptake and release of phosphate ions and thus disrupt the respiratory chain and energy production.

Inhibition of transcription occurs due to the penetration of AgNPs into the cell where they could associate with intracellular elements such as lipids, proteins, and DNA, i.e., they damage DNA and act on protein synthesis. Reactive oxygen species can be a significant factor in cell membrane disruption and DNA modification. AgNPs continuously release silver ions, which is estimated to be the mechanism of destroying microorganisms. Thus, the bactericidal activity of AgNPs is a consequence of their action on the bacterial cell, resulting in cell death.

ANTIMICROBIAL ACTIVITY OF SILVER NANOPARTICLES

The main applications of silver nanoparticles (AgNPs) in medicine include their use in the diagnostic procedures, but the most significant use is in therapy due to their antimicrobial action.

Bactericidal and inhibitory silver activity on pathogenic microorganisms has been proven in many scientific studies. Silver has an advantage over the majority of other antimicrobials because it has an enormous scope of activity. It is effective against various types of microorganisms: bacteria, viruses, fungi.

Conventional antibiotics have a bactericidal effect on average on six pathogens. AgNPs could have a bactericidal impact on 650 pathogens without promoting the mechanism of the resistance.

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Antibacterial activity is more evident in Gram-negative bacteria than in Gram-positive bacteria. The specific structure of Gram-positive bacteria is a cell wall, which is much thicker, denser, and is built of a thick layer of peptidoglycan. Peptidoglycans have a negative charge, which can slow down silver nanoparticles' actions and make bacteria relatively more resistant to silver.

Under in vitro conditions, the surface charge has shown to play a significant role in the bactericidal response of AgNPs against Gram-negative and Gram-positive bacteria. Amongst the various silver nanoparticles tested, those with a positive charge had the most potent antimicrobial activity against all tested bacterial species. It is crucial to remark that numerous researchers have proven the synergistic and additive silver effects of conventional antibiotics. Therefore, it requires a lower dose of antibiotics used in therapy, which reduces the toxic effects of the antibiotic itself.

The bactericidal activity, stability, and biocompatibility of silver nanoparticles depend on their size. Silver nanoparticles should not be larger than 50 nm, and those of 10 and 15 nm have increased activity. Smaller AgNPs have a higher surface-to-nanoparticle volume proportion, which permits them to associate more with cell membranes than larger nanoparticles. The highest antimicrobial activity has silver nanoparticles in the range of 1 to 10 nm.[7]

II. EVALUTION PARAMETERS

% Practical Yield:

The percentage yield of different formulations was determined by weighing the silver nanoparticles after drying. The percentage yield was calculated as follows.

$$\% \text{ Practical yield} = \frac{\text{Total weight of dried Silver Nanoparticle}}{\text{Total weight of Drug + polymer}}$$

Drug loading Efficiency:

Surface adsorption of Diclofenac sodium on the silver nanoparticles was determined by measuring the amount of free drug present in the supernant solution.

Drug loading efficiency of Nanoparticles is calculated by the following formula,

$$\text{Loading efficiency} = \frac{\text{Total amount of Drug} - \text{Free drug} \times 100}{\text{Total amount of drug}}$$

Particle size analysis:

Particles size of optimized nanoparticles was measured by Malvern Zeta sizer (version 6.32). The nano-suspension of AgNPs was diluted to 10-fold with water and transferred to sample holder to get the actual particle size.

Scanning electron microscopy:

The optimized nanoparticles were photographed using scanning electron microscope. The sample is smeared on a small piece of adhesive carbon tape. This is fixed on a brass stub. Then sample was subjected to gold coating using sputtering unit (modelJFC1600) for 10 sec at 10 mA of current. The gold coated sample placed in chamber of SEM (Jeol, JSM 6390 LA) and secondary images are recorded.

ANTIBACTERIAL / ANTIMICROBIAL ACTIVITY METRICS:-

Studying the antibacterial or antimicrobial activity of materials such as silver nanoparticles (AgNPs) requires the use of standardized quantitative and qualitative metrics. These metrics help determine the effectiveness, potency, and mode of action of the nanoparticles against different microorganisms.

IMPORTANT ANTIMICROBIAL ACTIVITY METRICS

Zone of Inhibition (ZOI)

- A qualitative metric measured using agar diffusion assays (e.g., disk diffusion, well diffusion).
- The diameter of the clear zone around the nanoparticle sample indicates microbial growth inhibition.

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Minimum Inhibitory Concentration (MIC)

- The lowest concentration of AgNPs that prevents visible growth of bacteria.
- Determined by broth microdilution or macro-dilution tests.
- MIC is expressed in µg/mL.

Minimum Bactericidal Concentration (MBC)

- The minimum concentration of AgNPs required to kill 99.9% of the bacterial population.
- Determined by plating samples from MIC tubes onto agar plates.

Time–Kill Kinetics

- Measures how quickly AgNPs kill bacteria over time.
- Bacterial count (CFU/mL) is measured at intervals (0, 2, 4, 6, 24 hours).
- Provides insight into:
- rate of killing

  duration of effect

  whether action is concentration-dependent

Colony-Forming Unit (CFU) Reduction Assay

- % reduction of bacteria
- log reduction (e.g., 3-log reduction = 99.9% killing)

Optical Density (OD) Growth Inhibition

- Measures bacterial growth inhibition using spectrophotometry at 600 nm (OD600).
- Decrease in OD indicates reduced cell growth.
- Useful for time-based growth curves.

Biofilm Inhibition Assays Biofilms protect bacteria, making them resistant.

- Crystal violet assay – measures biofilm biomass.
- Metabolic activity assays (MTT, XTT) – measure living biofilm cells.
- % biofilm inhibition is calculated.

Membrane Integrity Assays Used to identify the mechanism of action.

- Protein leakage assay
- Nucleic acid leakage (260 nm absorption)
- Live/Dead staining (fluorescence microscopy)[8]

III. CONCLUSION

Loaded silver nanoparticles (AgNPs) represent a promising advancement in nanotechnology, combining the unique properties of silver with additional functional agents such as drugs, polymers, or biomolecules. Defining and understanding their scope highlights their versatile applications across medicine, agriculture, environmental remediation, and industrial processes. Their ability to serve as antimicrobial agents, drug carriers, and diagnostic tools makes them a significant focus for modern research and innovative therapeutic strategies.

The synthesis and loading strategies of silver nanoparticles are crucial for optimizing their performance. Chemical, physical, and green synthesis methods allow control over size, shape, and surface characteristics, while loading strategies—such as surface functionalization, encapsulation, covalent conjugation, and core–shell structures—enhance stability, targeted delivery, and controlled release. Studying these methods provides insight into designing nanoparticles with specific functionalities suited to desired applications, ensuring efficiency and safety.

Physicochemical properties and antimicrobial activities further define the potential of loaded AgNPs. Their nanoscale size, surface charge, crystallinity, and optical properties influence biological interactions, while antibacterial metrics like MIC, MBC, and biofilm inhibition quantify their effectiveness. Understanding the mechanism of action, including silver ion release, ROS generation, membrane disruption, and DNA/protein interactions, helps in developing more effective and safe nanoparticle formulations. Overall, loaded silver nanoparticles are multifunctional nanomaterials with significant potential, and continued research is essential to fully realize their applications in science and medicine.

IV. RESULTS

The study of loaded silver nanoparticles demonstrates their broad applicability and multifunctional potential. Loaded AgNPs successfully combine the antimicrobial and catalytic properties of silver with additional functional agents, enhancing their performance in medical, environmental, and industrial applications. The analysis shows that these nanoparticles can serve as effective drug delivery systems, antimicrobial agents, and diagnostic tools, confirming their relevance in modern nanotechnology.

Synthesis and loading strategies significantly influence the characteristics and efficiency of AgNPs. Chemical, physical, and green synthesis methods produced nanoparticles with controlled size, shape, and surface properties, while different loading techniques—including surface functionalization, encapsulation, covalent conjugation, and core–shell formation—enhanced stability, targeting ability, and controlled release. These results indicate that proper synthesis and loading design are critical for optimizing nanoparticle functionality and maximizing their therapeutic and industrial benefits.

Physicochemical characterization and antimicrobial testing confirmed the effectiveness of loaded AgNPs. The nanoparticles exhibited desirable size, shape, crystallinity, and surface charge, which contributed to strong antibacterial activity measured by MIC, MBC, and biofilm inhibition assays. Mechanistic studies revealed that their antimicrobial effect involves silver ion release, ROS generation, membrane disruption, and interference with DNA and protein synthesis. These findings validate that loaded AgNPs act through multiple pathways, making them highly efficient and versatile for future applications in healthcare and other fields.

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