

A DISSERTATION ON
Green synthesis of Silver Nanoparticles by using *Albizia*
***Julibrissin* aqueous leaf extract**

SUBMITTED TO THE
DEPARTMENT OF BIOSCIENCES
INTEGRAL UNIVERSITY, LUCKNOW



IN PARTIAL FULFILLMENT
FOR THE
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IN BIOTECHNOLOGY
BY

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Enrollment no. 1700101894

M.Sc. Biotechnology (IV semester)

Department of Biosciences
Integral University, Lucknow

UNDER THE SUPERVISION OF

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TO WHOM IT MAY CONCERN

This is to certify that **Ms. Khadeeja Nezam**, a student of M.Sc. Biotechnology (IV semester), Integral University has completed her four months dissertation work entitled "*Green synthesis of Silver Nanoparticles by using Albizia Julibrissin aqueous leaf extract*" successfully. She has completed this work from 2 Feb to 2 June 2022 at the Department of Biosciences, Integral University, under the guidance of **Dr. Salman Khan**.

The dissertation was a compulsory part of her M.Sc. degree. I wish her good luck and a bright future.

(Dr. Snober S. Mir)

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June 2022

CERTIFICATE OF ORIGINAL WORK

This is to certify that the study conducted by **Ms. Khadeeja Nezam**, during the months 2 Feb to 2 June 2022 reported in the present thesis was under my guidance and supervision. The results reported by her are genuine and the script of the thesis has been written by the candidate herself. The thesis entitled "*Green synthesis of Silver Nanoparticles by using Albizia Julibrissin aqueous leaf extract*" is, therefore, being forwarded for acceptance in partial fulfillment of the requirements for the degree award of the student of M.Sc. Biotechnology (IV semester), Department of Biosciences, Integral University, Lucknow, (U.P).

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Khadeeja Nezam

Date

Nanotechnology

Nanotechnology is an emerging and energetic field of research and technology at the atomic, molecular, and macromolecular scales. Metal nanoparticles like gold, silver, platinum, zinc, and selenium receive greater attention from researchers due to their great properties (electronic photo-chemical and optical properties) [1]. Nanotechnology is rapidly becoming a ubiquitous technology with the potential to impact every aspect of modern human civilization. Almost every aspect of human endeavor will be affected, such as agriculture and food, communication, computer environmental monitoring, textiles, robotics, healthcare, and medical technology. More recently computer science has become-evolved in human-machine interface, artificial intelligence, and intelligent systems. The extensive use of computers and their wide application in the modern world have forced researchers to improve and manufacture smaller, faster, and more reliable computers. This objective can be fulfilled by nanotechnology. According to M.C. Roco, the third and fourth generation of nanotechnology would rely heavily on research in computer science.

Nanomaterials, controlled to nanocrystalline size (less than 100 nm), can show atom-like behaviors which result from higher surface energy due to their large surface area and wider band gap between valence and conduction band when they are divided to near-atomic size [2]. Nanoparticles comprise three layers.

- The surface layer may be responsible for small molecules, polymers, surfactants, and metal ions.
- The shell layer has a different chemically material from the core.
- The core is the central portion of the nanoparticles [3].

Nanotechnology is a relatively new branch of science that has found a wide range of applications that range from energy production to industrial production processes to biomedical applications.

Making anything to the nanotechnology scale for screening purposes presents enormous technical challenges. Manipulating matter at the nanometre scale is important for many electronic, chemical, and biological advances [4].

Nanotechnology applied to biological systems

1. Size is important

The main advantage of nanotechnology can control size according to resulting matter and devices.

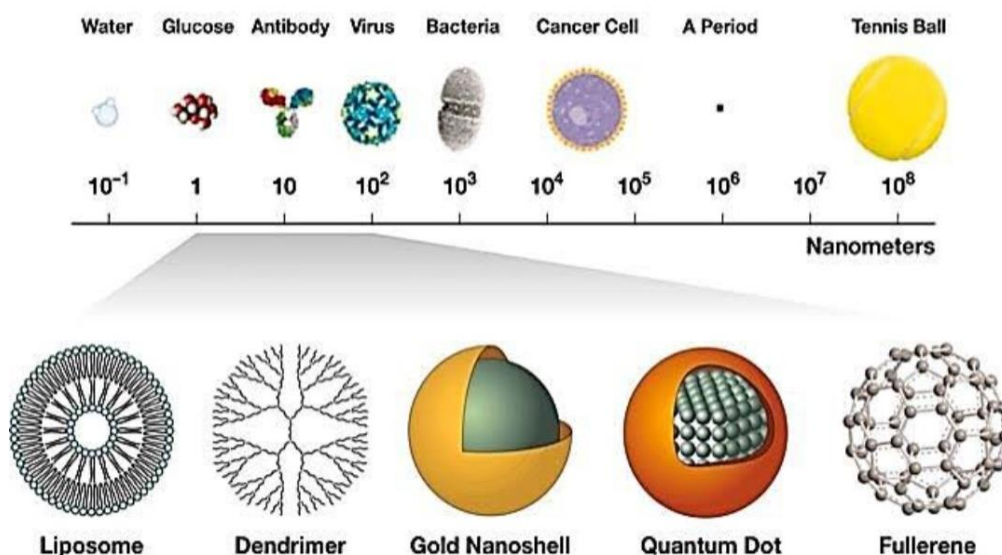


Fig. 1. The relative size of nanoparticles compared with a familiar item. Nanoscale constructs are smaller than human cells (10,000-20,000 nm in diameter) and organelles and similar in size to large biological macromolecules such as enzymes and receptors; hemoglobin, for example, is 5 nm in diameter, and the lipid bilayer surrounding cells is on the order of 6 nm thick. Nanoparticles smaller than 20 nm can transit through blood vessel walls. Magnetic nanoparticles, for instance, can image metastatic lesions in lymph nodes because of their ability to exit the systemic circulation through the permeable vascular epithelium [5].

2. Solubility matters

This advantage is perhaps most applicable in drug discovery and delivery. A common screening method during drug discovery and profiling is to score new compounds based on "Lipinski's Rule Of Five" [6].

"chemical leads" are based primarily on the compound's predicted solubility, as molecules with low solubility generally have poor absorption and decreased metabolic stability *in vivo* [7].

3. Targeting matters

In nanotechnology, Liposome is the earliest applied target in drug delivery. Size alone is not the only way that nanotechnology can enable targeting. Because of the highly tailorable surfaces of nanoscale constructs, it has become a relatively simple matter to "decorate" the nanoparticle surface with a variety of targeting agents. Resulting in targeted delivery of the drug [8].

Nanoparticles

Classification of nanoparticles

Nanoparticles are broadly classified into three classification

One dimension nanoparticles

One-dimensional system (thin film or manufactured surfaces) has been used for decades. Thin films (sizes 1-100 nm) or monolayer is now commonplace in the field of solar cells offering, different technological applications, such as chemical and biological sensors, information storage systems, magneto-optic and optical device, fiber-optic systems

Two-dimension nanoparticles

Carbon nanotubes

Three dimension nanoparticles

Dendrimers, Quantum Dots, Fullerenes (Carbon 60), (QDs) [9].

Silver nanoparticles

Silver, with the symbol Ag and atomic number 47, is a shining, extremely ductile, and malleable element that is somewhat harder than gold. It is one of the most fundamental components of our world. It can be found in nature as a native element, as an alloy with other metals (such as gold), and as minerals (e.g., chlorargyrite and argentite).

Silver has four different oxidation states chemically: Ag^0 , Ag^{1+} , Ag^{2+} , and Ag^{3+} , however, it is a chemically inert element that can be treated with nitric acid or hot concentrated sulfuric acid to produce soluble silver salts. Although it has outstanding heat and electricity conductivity, its applications in the electrical industry have been severely constrained because of its

higher cost. The metallic form of silver is water-insoluble, but its metallic salts, such as silver nitrate (AgNO_3) and silver chloride (AgCl), are. Its metallic salts have also been used to treat a variety of diseases and conditions, including epilepsy, gonorrhea, and gastroenteritis. Soluble silver compounds have a high absorptivity, which means they can have negative health effects if consumed in large amounts. Nonetheless, according to Chen and Schluesener (2008), silver is relatively non-toxic and non-carcinogenic to human key body systems such as the neurological, immunological, reproductive, and cardiovascular systems. As a result, silver demand has increased in recent years, primarily in the medical, plastics, and textiles industries. Silver has long been regarded as a safe and effective treatment for animal cells. Because silver is non-toxic to animal cells, it has long been thought to be a safe and effective antibacterial metal, particularly because it is very poisonous to bacteria like *Escherichia coli* (*E. coli*) and *Staphylococcus aureus*. As a result, silver-based compounds have attracted a lot of attention as antimicrobial agents in applications like burn care for centuries. Antibiotic resistance has become a driving force in the search for silver nanoparticle production in recent years, owing to the rising threat of drug resistance produced by antibiotic overuse [10].

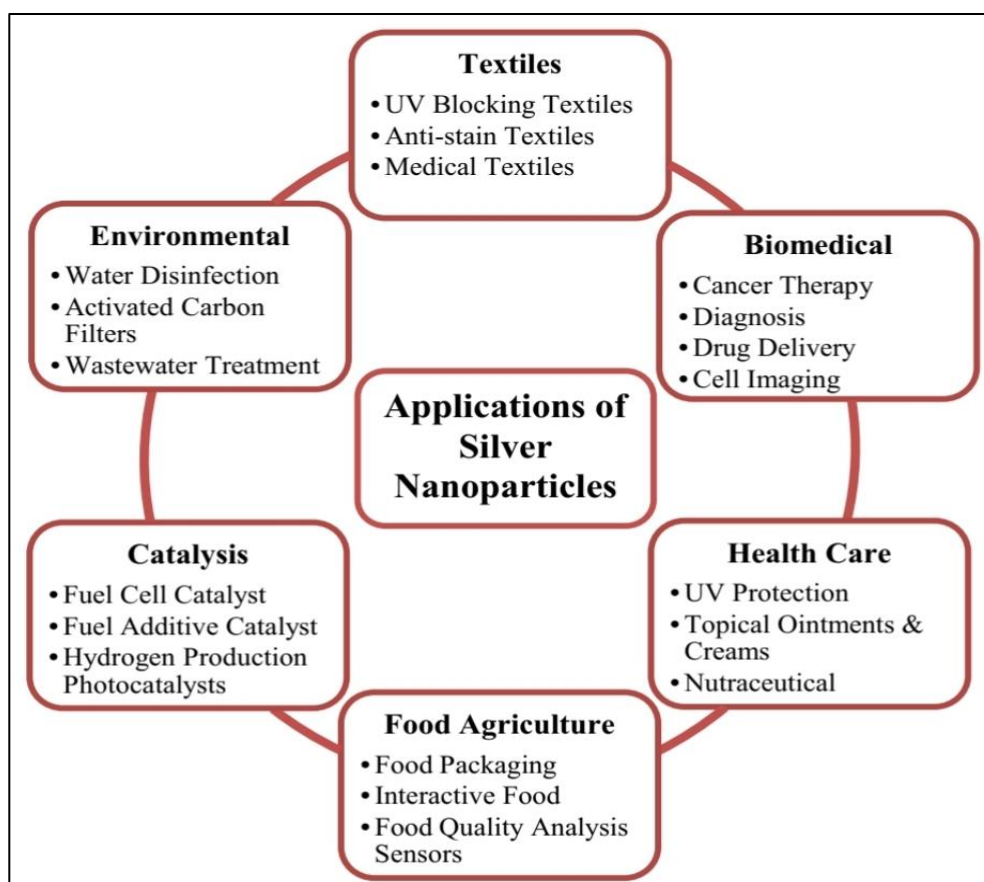


Fig. 2. Applications of silver nanoparticles.

***Albizia julibrissin* (Persian silk tree, pink silk tree)**

Albizia julibrissin Persian silk tree, pink silk tree is a Fabaceae tree species that is commonly referred to as "silk tree" or "mimosa" in the United States. It is commonly used as a decorative plant in parks and gardens in India, where it is prized for its leaf texture and blossoms. *Albizia julibrissin* flower extract now has antidepressant, antidiabetic, anti-inflammatory, and anti-obesity characteristics, as well as a reduction in both its spontaneous and passive activities. It's also a well-known insomnia cure in Chinese medicine, and it has an essential and strong anti-tumor effect in vitro. As a result, its essential oil and extract are commonly used to treat dysphoria-induced wakefulness, depression, forgetfulness, and dreaminess. *Albizia julibrissin's* pharmacological activity can be used to identify proof of these extraordinary *Albizia julibrissin* effects. Although not much of this dimension has been investigated, *Albizia julibrissin* can be used for future research and can be of tremendous therapeutic utility. *Albizia julibrissin* is a

traditionally used drug for snake bites and leprosy. In traditional Indian and Chinese medicine, *Albizia* plants are used therapeutically for insomnia, irritability, and wounds, as anti-dysenteric, antiseptic, antitubercular, etc [11].

Review of literature

Nanotechnology

Nanotechnology is one of the most promising 21st-century technologies. It is the ability to see, measure, manipulate, assemble, control, and manufacture materials at the nanoscale scale to put nanoscience theory into practice. Nanotechnology is defined as "a science, engineering, and technology conducted at the nanoscale (1 to 100 nm), where unique phenomena enable novel applications in a wide range of fields, from chemistry, physics, and biology, to medicine, engineering, and electronics" by the National Nanotechnology Initiative (NNI) in the United States. In 1959, Nobel Laureate and American physicist Richard Feynman introduced the concept of nanotechnology. Feynman gave a talk titled "There's Plenty of Room at the Bottom" at the California Institute of Technology during the American Physical Society's annual meeting (Caltech). Feynman proposed the hypothesis "Why can't we write the full 24 volumes of the Encyclopedia Britannica on the top of a pin?" and detailed a vision of utilizing machines to build smaller machines and down to the molecular level. This new concept indicated that Feynman's hypotheses were valid, and he is thus regarded as the founder of modern nanotechnology. After fifteen years, Norio Taniguchi, a Japanese scientist, was the first to adopt and define the word "nanotechnology" in 1974. After Feynman discovered this new field of inquiry, which piqued the curiosity of many scientists, two methodologies were created to describe the various options for nanostructure production. There are two types of manufacturing approaches: top-down and bottom-up, which differ in terms of quality, speed, and cost [12]. Another Japanese scientist, Iijima, developed carbon nanotubes, which advanced nanotechnology even further.

The developing fields of nanoscience and nanotechnology sparked significant attention at the turn of the century. Feynman's fame in the United States, as well as his concept of manipulating matter at the atomic level, influenced national research agendas. During a speech at Caltech on January 21, 2000, President Bill Clinton called for financing of study in this new technology. President George W. Bush signed the 21st Century Nanotechnology Research and Development Act into law three years later. The Act established the National Technology Initiative and made

nanotechnology research a national priority (NNI) [13]. Nanotechnology, or the creation of systems and devices at the molecular level, is a multidisciplinary scientific discipline that is rapidly evolving. The promise of breakthrough advances in health, communications, genetics, and robotics sparked the development of nanotechnology. Miniaturization appears to provide cost-effective and faster-functioning mechanical, chemical, and biological components. Nanoscale things, on the other hand, have extraordinary self-ordering and assembly behaviors under the influence of pressures that are fundamentally distinct from macroscale objects. Nanotechnology is made possible by these distinct behaviors, and extending our understanding of these processes will undoubtedly lead to new approaches to improving human life quality. A comprehensive list of nanotechnology's prospective uses is impossible to compile, but one of nanotechnology's greatest benefits will undoubtedly be in the development of novel and effective medical treatments (i.e., nanomedicine)[14]. Nanotechnology the creation and utilization of materials, devices, and systems through the control of matter on the nanometer has been applied to molecular diagnostics. Nanotechnologies enable the diagnosis at the single-cell and molecule level and some of these can be incorporated into the current molecular diagnostics such as biochips [15].

Nanotechnology In Nanomedicine

Nanomedicine is a subdivision of nanotechnology. The medical application of nanotechnology is what is meant by Nanomedicine. The term nano means mini scale, extremely small, hence, Nanomedicine deals with drug formulations that have sizes in the nano range. Devices built with the help of nanotechnology are used in the monitoring, repair, construction, and control of human biological systems. Having a large surface area, the nanoparticles can be attached to drugs, and proteins, targeting moieties and diagnosing agents, making them useful in medicine. Nanotechnology is helping to realize mankind's dream of fighting against many complex illnesses like cancer, multiple sclerosis, cardiovascular diseases, Alzheimer's and Parkinson's diseases, diabetes as well as some inflammatory or infectious diseases (like AIDS) [16].

Nanomedicine's goal can be broadly defined as the total monitoring, control, building, repair, defense, and improvement of all human biological systems, starting at the molecular level and progressing through the use of engineered devices and nanostructures to accomplish medicinal benefits. Nanoscale should be understood to encompass active components or objects with sizes ranging from one nanometre to hundreds of nanometres in this context. These could be used in a micro-device (with a macro interface) or a biological setting. However, nano-interactions within the context of a bigger device or directly within a sub-cellular (or cellular) system are always the focus.

Nanosciences and nanotechnologies are the study and manipulation of materials on an extremely small scale. A single human hair is around 80,000 nanometres thick. As a result, nanomedicine works on the same scale as biological molecules and structures within living cells, which is roughly 100 nanometres or less. Proteins range in size from 3 to 10 nanometres (nm), while red blood cells are between 6000 and 8000 nm in size. The nanoparticulate systems range in size from a few nanometres, such as micelles, to hundreds of nanometres, such as liposomes. Drug delivery systems, for example, can easily interact with biomolecules on both the cell surface and within.

The nanoparticulate systems range in size from a few nanometres, such as micelles, to hundreds of nanometres, such as liposomes. Drug delivery systems, for example, can easily interact with biomolecules on both the cell surface and within. As a result, nano-drug delivery systems may not only transport encapsulated or grafted tiny chemotherapeutic medicines with a diameter of less than a dozen nanometres but also deliver them inside cells once they have pierced them. Such systems can also be coated with antibody fragments on their surface to target specific organs, enhancing drug delivery selectivity.

The ability to work at the same size of various biological processes, cellular systems and organic molecules has fueled nanotechnology's success in the healthcare industry; as a result, medicine has viewed nanotechnology as the ideal option for the detection and treatment of many disorders. The topic of drug distribution is one of the many uses of nanotechnology in the

medical field. The development of protocols and methods for the synthesis, functionalization, and application of nanoparticles and nanocarriers has flooded the scientific and clinical communities with new therapeutic approaches ranging from molecular targeting to radiofrequency ablation, and personalized therapies to minimally invasive techniques.

Nanoparticles

Nanoparticles are particulate dispersions or solid particles having a size between 10 and 1000 nanometers. A nanoparticle matrix is used to entrap, encapsulate, or attach the medication. Nanoparticles, nanospheres, and nanocapsules can be made depending on the method of preparation. Nanocapsules are matrix systems in which the drug is physically and uniformly spread, while nanospheres are cavity systems in which the drug is contained in a cavity enclosed by a unique polymer membrane. Biodegradable polymeric nanoparticles, particularly those coated with hydrophilic polymers such as poly(ethylene glycol) (PEG) known as long-circulating particles, have been used as potential drug delivery devices in recent years due to their ability to circulate for long periods, target a specific organ, and deliver proteins, peptides, and genes [17].

Controlling particle size, surface characteristics, and release of pharmacologically active substances to act as the site-specific action of the drug at the therapeutically optimal rate and dose regimen are the key goals in designing nanoparticles as a delivery system. Even though liposomes have been used as potential carriers with unique advantages such as protecting drugs from degradation, targeting the site of action, and reducing toxicity or side effects, their applications are limited due to inherent problems such as low encapsulation efficiency, rapid leakage of water-soluble drugs in the presence of blood components, and poor storage stability. Polymeric nanoparticles, on the other hand, have certain distinct advantages over liposomes. For example, they can aid boost drug/protein stability and provide valuable controlled release features [18].

Classification of nanoparticles

The morphology, size, and form of nanoparticles are used to classify them

into different categories. This overview discusses some of the most important nanoparticle classes.

1. **Organic nanoparticles** are non-toxic and biodegradable, and certain organic nanoparticles, such as micelles and Liposomes, feature a hollow sphere. It is also known as the name of heat and light-sensitive nanocapsule. Because of these features, organic nanoparticles are an excellent alternative for medication delivery. Nanoparticles are also commonly used in the delivery of target drugs. Polymeric nanoparticles are another name for organic nanoparticles. The nanosphere or nanocapsule is the most well-known shape of organic or polymeric nanoparticles. The matrix particles have a solid overall mass and adsorb other molecules at the
2. **Liposomes** were the first NP platform. Liposomes were first proposed as a model of biological membranes in 1965. Liposomes have progressed from being a model in biophysical study to become one of the first NP platforms to be used for gene and drug delivery. Liposomes are spherical vesicles that self-assemble in aqueous systems and comprise a single or multiple bilayered lipid structure. Liposomes have a wide range of compositions, as well as the capacity to carry and protect a wide range of biomolecules, as well as biocompatibility and biodegradability. Liposomes have been well-studied and widely used as transfection agents of genetic material into cells (lipofection) in biology studies due to these advantages. In most cases, a cationic lipid is used to build an aggregation with anionic genetic material. Liposomes can also be used as therapeutic carriers since their design allows for the trapping of hydrophilic substances inside the core and hydrophobic medicines within the lipid bilayer. Liposomes have been coupled with biocompatible polymers such as polyethylene glycol to improve their circulation half-life and stability in vivo (PEG). Targeting ligands can be added to liposomes to boost the accumulation of diagnostic and therapeutic substances within desired cells. There are now twelve liposome-based medicinal medicines in clinical trials [20].
3. **Dendrimers** are another form of polymeric NP. Dendrimers are regularly branching macromolecules produced from amino acids, sugars, and

nucleotides, which can be synthetic or natural. They have a central core, inner branch layers, and a surface on the outside. Dendrimers with well-defined size, shape, and branching length/density can be made using a variety of these components. Dendrimers can be used as sensors as well as medication and gene delivery vehicles due to their distinctive architecture.

Chemical linkage, hydrogen bonding, and/or hydrophobic interaction can all be used to load tiny molecules into the cavities of dendrimers' cores. Chemical functional groups for molecular targeting groups, detecting and imaging agents, and medicinal attachment sites can all be easily produced on the outer surface [21].

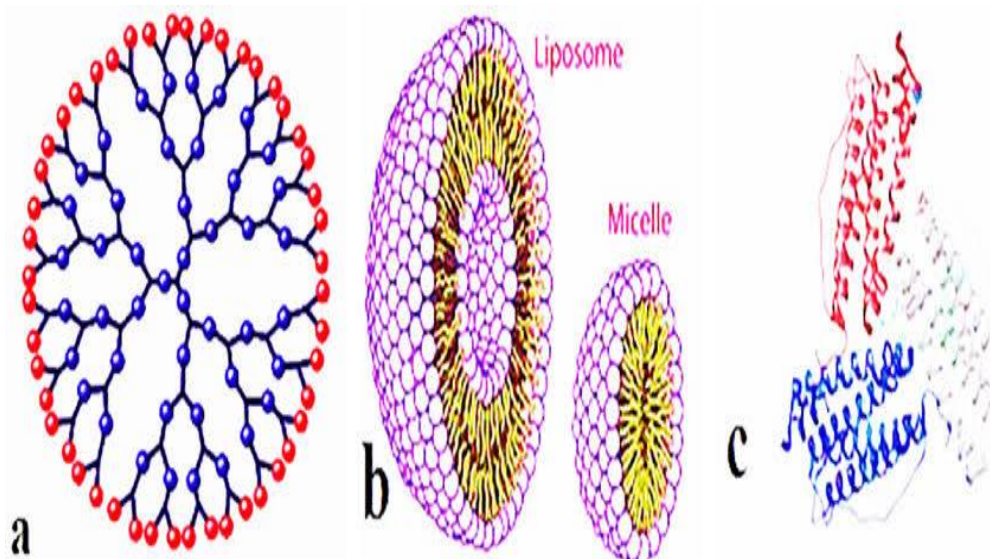


Fig. 3. Organic nanoparticles (a) Dendrimers (b) Liposomes and Micelles (c) Ferritin.

Inorganic nanoparticle

Inorganic nanoparticles do not contain carbon. Inorganic nanoparticles are not poisonous. Biocompatible and hydrophilic inorganic nanoparticles Organic nanoparticles are less stable than inorganic nanoparticles.

Metal and metal oxide nanoparticles are two types of inorganic nanoparticles.

Metal nanoparticles

Metallica nanoparticles are made from metals using destructive or

constructive processes. Pure metal nanoparticles are created using metal precursors. Due to plasma resonance characteristics, metal nanoparticles have unique optoelectrical capabilities. Shape, facet, and size all influence the production of metal nanoparticles. All metal nanoparticles can be manufactured [22].

Nanoparticles like aluminum, zinc, gold, iron, lead, silver, zinc, etc

Metal oxide nanoparticles

The goal of metal oxide nanoparticle production is to change the properties of their corresponding metal nanoparticles, such as turning iron nanoparticles into iron oxide nanoparticles. When compared to iron nanoparticles, the reactivity of iron oxide nanoparticles is higher. Metal oxide nanoparticles are generated due to an increase in reactivity and efficiency of metal oxide. Zinc oxide, silicon dioxide, iron oxide, aluminum oxide, cerium oxide, titanium oxide, and magnetite are examples of metal oxide nanoparticles.

Ceramic nanoparticles

Nonmetallic solids are another name for ceramic nanoparticles. Ceramic nanoparticles are made by heating or cooling them repeatedly. Polycrystalline, amorphous, porous, dense, or hollow ceramic nanoparticles are possible. The researchers focused on these nanoparticles because of their wide range of uses, including dye photodegradation, photocatalysis, catalysis, and imaging.

Biological nanoparticles or bio nanoparticles

An assemblage of atoms or molecules generated in a biological system with at least one dimension in the range of 1–100 nm is known as a biological or bio-nanoparticle. Bionanoparticles are nanoparticles that exist in nature. These nanoparticles are classified as having an intracellular or extracellular structure.

Lipoproteins and viruses are examples of extracellular structure, while magnetosomes are an example of intracellular structure. Bionanoparticles include magnetosomes, exosomes, ferritin, lipoproteins, and viruses.

Nanomaterials

Nanomaterials are materials with at least one structural component or unit that is smaller than one micrometer in size. Nanomaterials are matter's building blocks (less than 0.2 nm).

Carbon-based nanomaterials

Carbon-based nanomaterials are made entirely of carbon. Carbon-based nanoparticles include fullerenes, carbon nanotubes (CNT), graphene, carbon black, and carbon nanofibers.

Fullerenes

C₆₀ or C₇₀ fullerenes are possible. The fullerenes are made up of hollow cages of nanomaterials. It gains economic attention due to its electrical conductivity, electron affinity, structure, strength, and adaptability. Fullerenes are made up of carbon units that are pentagonal or hexagonal. Fullerenes have sp² hybridized carbon atoms that connect them. Fullerenes constructed of C₆₀ or C₇₀ have diameters of 7.114 and 7.648 nm, respectively. A single layer or multilayer fullerene can exist.

Carbon nanotubes (CNT)

Carbon Nano Tubes are elongated tubular structures with a diameter of 1–2 nm. Based on diameter, a carbon nanotube can be classified as semiconducting or metallic. CNT has a structure that looks like a graphite sheet rolling on itself. Single-walled (SWNTs), double-walled (DWNTs), and multi-walled CNTs are classified based on their rolling properties (MWNTs).

Graphene

Carbon in the form of graphene is allotropic. It's a two-dimensional hexagonal honeycomb carbon atom lattice with a planar surface. Graphene is a 1-nanometer thick carbon nanotube. The same graphene nano foil is transferred into carbon nanofiber as carbon nanotubes, however, instead of elongating cylindrical tubes, the nano foil is twisted into a cup or cone.

Carbon black

It is a carbon-based amorphous substance. Carbon black has a spherical form. The diameter varies between 20 and 70 nanometers. Because of the

high contact between the particles, they are bound in an aggregate.

Classification of nanomaterial-based on the dimension

Nanomaterials are classified into dimension (1D) nanomaterials, two-dimension (2d) nanomaterials, and three Dimension (3D) nanomaterials.

One-dimension nanomaterial

The number 10^9 appears in the name Nano, which represents one-billionth of any unit that results in the development of one-dimensional nanomaterials such as thin films. Nanomaterials offer a wide range of uses in chemistry, pharmaceuticals, electronics, and engineering. The monolayers or thin films range in size from 1 to 100 nanometers. These nanomaterials are very important in research and are used in the fabrication of electronics, storage systems, nanoscale LEDs, chemical and biosensing magnetooptics, fiber optic systems, and optical devices. one dimension of nanomaterial play role in the construction of important materials at nanoscales such as nanotubes, a nanobelt, nanowires, nano-ribbons, and hierarchical nanostructures.

Two-dimension nanomaterial

2D nanostructures have two dimensions that are outside of the nanometric size range and have a unique shape. 2D nanomaterials are used as building blocks for key components of nanodevices Nanocontainers, sensor photocatalysts, nanoreactors, and templates for 2D structures are all examples of two-dimensional nanomaterials. Two-dimensional nanoparticles include carbon nanotubes.

Three-dimension nanomaterial

The behavior of nanomaterials is determined by the shape size dimension and morphology, which are the fundamental parameters for nanostructure application and performance. Three-dimensional nanomaterials have piqued interest in medical science and research throughout the last decade. These nanoparticles have a wide range of uses in catalysis, batteries, and reactant and product transport in magnetic materials. Fullerenes, Dendrimers, and Quantum dots are examples of three-dimensional

nanoparticles

Synthesis of nanoparticle

Nanoparticles can be synthesized using a variety of processes, which are divided into two categories. i.e

1. Top-down synthesis

The destructive approach is used in this synthesis. The larger molecule (bulk material) is broken down into smaller molecules, which are then transformed into nanoparticles. Top-down synthesis methods include grinding or milling, physical vapor deposition, and other damaging methods.

- (i) Thermal decomposition method:** It's an endothermic process in which heat causes chemical degradation. This heat breaks the chemical link in the molecule. The decomposition temperature can be defined as the temperature at which a chemical reaction occurs. The nanoparticles are the result of meta breakdown at a certain temperature.
- (ii) Mechanical method/ball-milling method:** It is a low-cost approach for making nanoparticles from bulk. The simplest mechanical method is ball milling. Ball milling produces nanoparticles by attrition. It's the process of transferring kinetic energy from the grinding medium to the substance being reduced. Consolidation and compaction, an industrial-scale process in which nanoparticles are "put back together," are used to create material with improved characteristics. This method is used to create various metal alloys.
- (iii) Lithographic methods** Are top-down methods capable of producing micron-sized features for the most part, although they are energy-intensive and require expensive equipment. For numerous decades, lithography has been employed to create printed circuits and computers. Nanoimprint lithography is a distinct sort of lithography from traditional lithography. It's a lot like template synthesis. A template is created first, and then a soft polymeric material is stamped to make the pattern. Stamped material is created via the top-down process. Nanosphere lithography uses latex spheres to create templated matrices. Photolithography, electron beam

lithography, soft lithography, focussed ion lithography, nano-imprint lithography, and dip pin lithography are examples of lithography processes. Contact and proximity printing, as well as projection printing, were all part of photo-lithography.

(iv) Laser ablation: Simple strategies for the synthesis of nanoparticles from various solvents include laser ablation synthesis in solution. Irradiation of various metals submerged in solution with a laser beam condenses a plasma, resulting in nanoparticles. It's a highly effective top-down way of reducing metal to nanoparticles that's different from traditional chemical approaches. Laser ablation techniques are used to create stable nanoparticles that do not require any stabilizing agents or chemicals.

(v) Sputtering: It is a phenomenon in which nanoparticles are deposited by ejecting particles from it. Annealing is a great way to deposit a thin coating of nanoparticles. The size and shape of nanoparticles are determined by the following factors mention

- (i) Temperature
- (ii) The thickness of the layer
- (iii) Duration of annealing
- (iv) Substrate etc

Bottom-up method

The constructive method is also known as the bottom-up method. It's the inverse of the top-down approach. In this method, nanoparticles are formed from relatively simpler substances. The bottom-up method includes Chemical vapor deposition (CVD), Sol-gel, spinning, pyrolysis, and biological synthesis.

Chemical vapor deposition (CVD) method

It's a technique for depositing a thin coating of the gaseous reactant on a substrate. A reaction chamber is used to deposit the thin film. A chemical reaction occurs when gas comes into contact with a heated substrate. As a result of this reaction, a thin coating of the product is formed on the substrate's surface. This thin film is saved and put to good use. The CVD process produces hard, robust, homogeneous, and very pure nanoparticles. The CVD process has several drawbacks, including the

need for specialized equipment and the generation of very poisonous gaseous by-products.

Sol-gel method

The words sol-gel are a combination of the phrases sol and gel. Sol is a colloid made up of suspended solid particles in a continuous liquid. A gel is a solid macromolecule that dissolves in a liquid medium. The sol-gel method is the most popular bottom-up method for nanoparticle synthesis due to its simplicity. It's a procedure in which an appropriate chemical solution serves as a catalyst. Metal oxide and chloride are common precursors in the sol-gel technique. The precursor is distributed in the host liquid using various processes such as stirring, sonication, and shaking. To recover the nanoparticles, the resultant solution is separated into a solid phase and a liquid phase using various procedures such as filtration, sedimentation, and centrifugation.

Spinning

Spinning is used to make nanoparticles. The nanoparticles are created using a spin(SDR) device, which consists of a revolving disc with physical parameters such as temperature that can be adjusted. The reactor is filled with nitrogen or inert gases to prevent chemical reactions and remove oxygen. Inside the chamber or reactor, liquids such as water and precursor are pumped. Disc surface, liquid/precursor ration, disc rotation speed, liquid flow rate, and feed location all influence the properties of nanoparticles synthesized from SDR.

Pyrolysis

Pyrolysis is the most popular commercial method for making nanoparticles. In this procedure, the precursor is burned with a flame. The precursor could be a liquid or a vapor. The precursor is placed in a high-pressure furnace to retrieve nanoparticles. When a high temperature is required, a laser or plasma may be used instead of a flame. Evaporation is facilitated by the high temperature.

Biological synthesis

Plant extract and microorganisms such as bacteria and fungi are used in biological synthesis to create nanoparticles. Phytonanotechnology has demonstrated a new field for nanoparticle production that is eco-friendly, easy, and cost-effective. The advantages of phyto-nanotechnology include scalability, biocompatibility, and nanoparticle synthesis using a universal solvent (water) as a reducing agent. Plants are used in phyto-nanotechnology to make nanoparticles. Plant parts such as the root, fruit, stem, seed, and leaf are used to make nanoparticles. The actual mechanism of plant-based nanoparticle creation is yet unknown. Organic acids, proteins, vitamins, and secondary metabolites such as alkaloids, flavonoids, terpenoids, polysaccharides, and heterocyclic compounds are responsible for the formation of diverse types of nanoparticles.

Microorganisms are considered nano factories, and they have a lot of potential as a cost-effective, environmentally friendly, and energy-efficient technique for the synthesis of nanoparticles. The presence of several reductase enzymes in the microbe, allows it to collect and detoxify heavy metals. These reductase enzymes are crucial in the transformation of metal salts into nanoparticles. Various microorganisms such as yeast, fungi, and bacteria have been discovered in the last few years. As a reducing and capping agent, proteins, reducing cofactors, metal resistance genes, enzymes, and organic materials play a key role in the creation of nanoparticles.

Advantages of green or biological synthesise

- Energy Efficient
- Low-cost production
- Fewer
- Safe product
- Economical
- Lesser waste, therefore, is also called an environmentally friendly
- Competitive advantages
- Protect human health and communities
- Use in the pharmaceutical industry and other biomedical applications.

Plant extract: To decrease and stabilize nanoparticles, many plants can be employed. Many researchers have employed a biological method to synthesize metal or metal oxide nanoparticles by utilizing various plant parts such as the leaf, stem, root, and fruit

Plans include a variety of biomolecules that reduce metal salts to nanoparticles, including proteins, coenzymes, and carbohydrates.

Bacteria: Bacteria can decrease metal ions and are consequently used in the production of nanoparticles. Metallic and other new nanoparticles are made using a range of bacterial species.

Fungi: The use of fungus in the biological synthesis of metal or metal oxide nanoparticles is also a very efficient approach with a well-defined shape. Fungi operate as a biological agent for nanoparticle formation due to the presence of intracellular enzymes. Bacteria generate fewer nanoparticles than fungus.

Yeast: Yeast are single-celled creatures

.A total of 1500 yeast species have been identified. Several scientists have reported on the synthesis of nanoparticles using yeast. Yeast is used to making a large number of nanoparticles or nanoparticles [23].

Characterization Of Nanoparticles

The term "characterization" in material science refers to the broad and generic techniques used to investigate a material's properties and structure.

For a scientific understanding of the material, this fundamental process is required. Any process that deals with material analysis, such as mechanical testing, thermal analysis, and density computation, falls under the category of characterization. Some techniques are qualitative, while others are quantitative.

Scanning Electron Microscopy (SEM)

With the direct observation of the nanoparticles, this electron microscopy-based approach evaluates their size, shape, and surface morphology. As a result, scanning electron microscopy has several advantages when it comes to morphological and size examination.

They do, however, provide limited information on the population size distribution and genuine average. The solution of nanoparticles should first be transformed into a dry powder before SEM characterization. This dry powder is then deposited on a sample holder before being sputter-coated with a conducting metal (for example, gold). After that, the entire sample is scanned with a concentrated electron beam. Surface characteristics are determined by secondary electrons released from the sample surface.

The polymer of the nanoparticles, which must be able to tolerate a vacuum, is frequently damaged by this electron beam.

SEM measurements of average mean size are analogous to results from dynamic light scattering. Furthermore, these procedures are time-consuming, expensive, and typically require additional information about sizing distribution

Applications

When compared to a light microscope, the SEM produces much more detailed three-dimensional images at much higher magnifications (up to 300000). The photos are black and white because they were made without the use of light waves. SEM can scan the surface structure of polymer nanocomposites, fracture surfaces, nanofibres, nanoparticles, and nanocoatings.

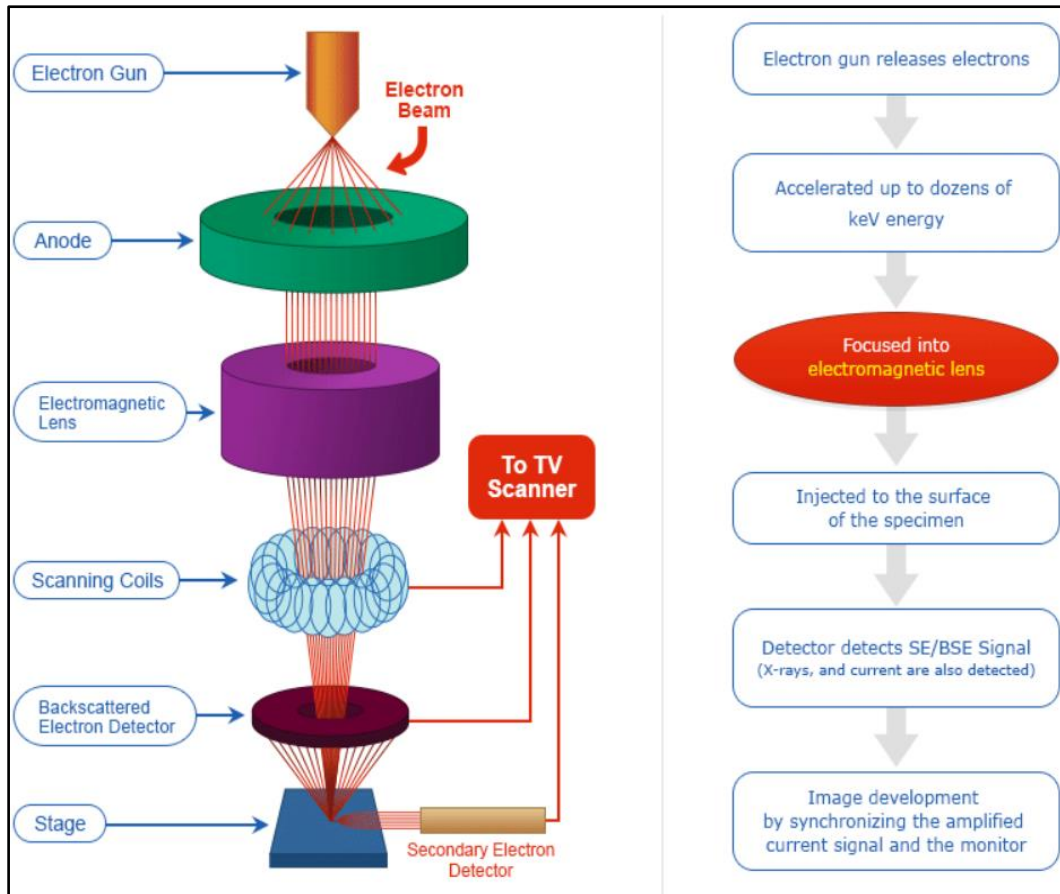


Fig. 4. Schematic diagram on the working of SEM.

Transmission Electron Microscopy (TEM)

Transmission electron microscopy is a type of microscopy in which a stream of electrons is passed through an ultra-thin material and reacts as it travels through it. The electrons passing through the specimen generate a picture.

An objective lens magnifies and focuses the image, which then displays on an imaging screen.

Applications

The transmission electron microscope (TEM) is widely utilized in material science and metallurgy, as well as biological sciences. The specimens in both circumstances must be extremely thin and able to endure the high vacuum inside the instrument. The maximum specimen thickness for biological specimens is about 1 millimeter.

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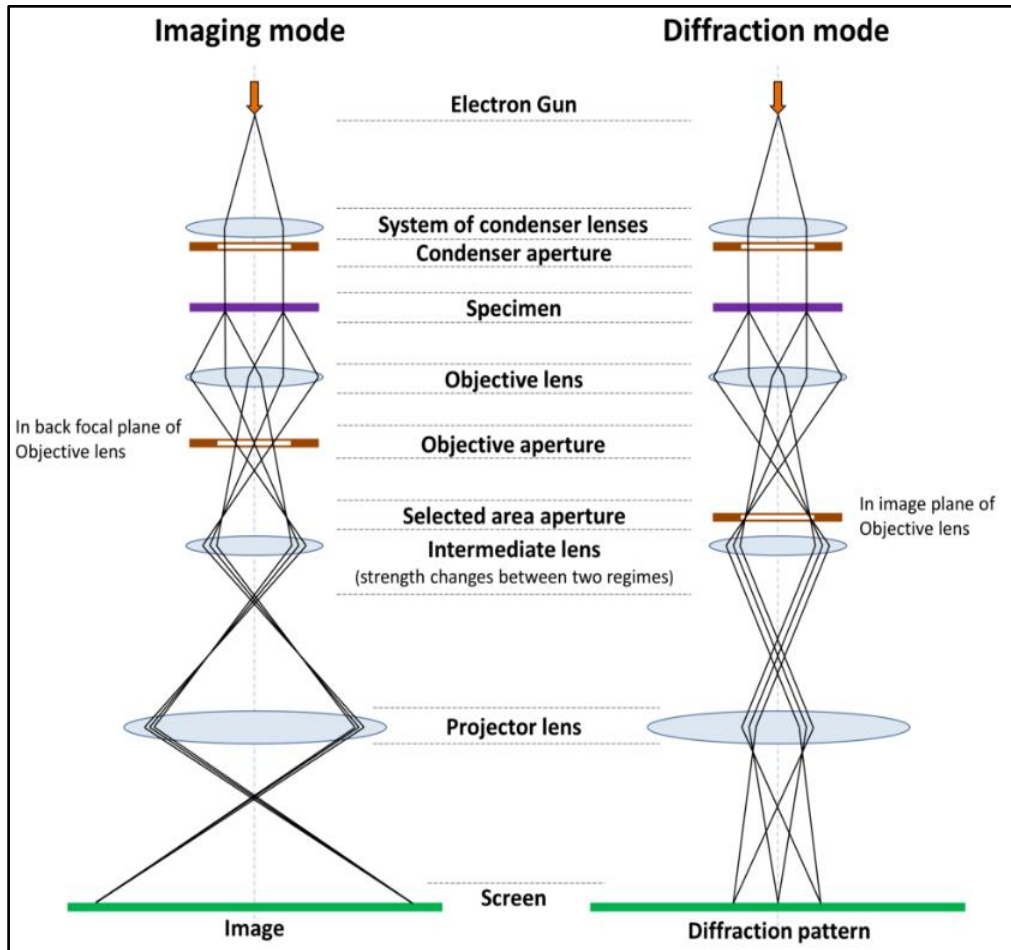


Fig. 5. The basic component of TEM

Biological specimens are often kept at liquid nitrogen temperatures after embedding in vitreous ice, or fixed using a negative staining agent such as uranyl acetate or by plastic embedding to endure

Ultraviolet-visible (UVs-vis) spectroscopy

A light source, reference and sample beams, a monochromator, and a detector make up an ultraviolet spectrophotometer. A sample of a substance is exposed to UV light from a light source, such as a Xenon lamp, to acquire its ultraviolet spectrum.

Application

The plasmon resonance is responsible for the distinctive and extraordinary

optical phenomena of some metals, such as silver and gold. Because of a collective resonance of the conduction electrons in the metal known as surface plasmon resonance, metallic (silver or gold) nanoparticles, generally, 40–100 nm in diameter, scatter optical light elastically with amazing efficiency. These plasmon resonant nanoparticles display the surface plasmon resonance peak in UV absorption spectra. The Plasmon resonance's magnitude, peak wavelength, and spectral bandwidth are all affected by the particle's size, shape, and material composition, as well as the surrounding environment.

Fourier transform infrared spectroscopy (FTIR)

FTIR analysis is a technique for identifying organic, inorganic, and polymeric materials by scanning them with infrared light. A change in the material composition is indicated by changes in the absorption band pattern. FTIR can be used to identify and characterize unknown components, discover impurities in a substance, locate additives, and detect decomposition and oxidation.

Dynamic light scattering (DLS)

The DLS technique is used to characterize colloidal fluids and nanoparticles.

DLS measures the scattered light from a laser as it moves through a colloidal solution. The modulation of the intensity of scattered light as a function of time is studied, and the particle size can be calculated using this information.

Zeta potential (ZP)

The zeta potential is a measurement of the effective electric charge on the nanoparticle's surface that quantifies the colloidal particles' charge stability. When a nanoparticle has a net surface charge, a greater concentration of oppositely charged ions near the nanoparticle's surface screens the charge. The ion layer moves with the nanoparticle, forming an electrical double layer consisting of the surface charge layer and the oppositely charged ions. The zeta potential is the difference in potential between the fluid layer containing oppositely charged ions and the bulk fluid in which the

particle is suspended. Negatively charged particles will bond to positively charged surfaces, and vice versa. The zeta potential's magnitude reveals information about the particle's stability. The greater the magnitude, the more stable the system is due to stronger electrostatic repulsion.

- In the 0–5mV range, particles tend to aggregate.
- Minimally stable particles have a voltage range of 5–20mV.
- Highly stable particles have a voltage range of 40+mV, whereas moderately stable particles have a voltage range of 20–40mV.

Another major factor is the pH of the solution at a certain pH, the surface charge can be brought to zero, known as the isoelectric point.

Applications

Nanotechnology applications in the biology field;

Applications in medicine

Nanoparticles have a wide range of medical applications, including:

- Fluorescent biological labeling
- Drugs and gene delivery
- Some of the applications. Useful in Pathogen Biological Detection
- Useful in Protein Detection
- For DNA structure probing
- For tissue and cell engineering
- Used to destroy tumors by heating them (hyperthermia)
- It's useful for separating and purifying biological cells and molecules
- Improvements in MRI
- Many types of biosensors based on nanoparticles are used to diagnose various diseases.
- Cellular imaging in Phagokinetic investigations

The best property of nanoparticles is surface plasmon resonance, which can be seen in metal nanoparticles. The scattering intensity of nanoparticles, on the other hand, increases as the number of particles increases. The addition of this property allows the specific molecule to be used in a variety of expected applications in living biological tissue by covering the surface of a cancer cell, for example, it is now possible to

distinguish between healthy and malignant cells. It is accomplished by the presence of antibodies, which must be united by the gold nanoparticle. Even though the gold nanoparticle-antibody junction is adequately dispersed in the healthy cell.

Application of nanoparticles in food and agriculture

Nanotechnology has brought new water filtration and desalination processes, but they will be more cost-effective. The food business can also benefit from nanotechnology. For example, nanotechnology can be used to create novel functional materials and equipment for food preservation and biosecurity. With the use of nanotechnology, the Bayer Company developed airtight plastic packaging. This plastic packaging keeps food fresh. Genetic alterations in the agricultural plant's constitution can be made using nanotechnology.

Application as a chemical catalyst

Nickel, lead, silver, and platinum have all been used as unique metal catalysts in chemical processes. The dissociative adsorption of hydrogen and oxygen molecules on the surface of the gold is not possible below 200 degrees Celsius. Because gold nanoparticles are not reactive during hydrogenation and oxidation reactions, gold material is utilized as a catalyst. Haruta, on the other hand, observed that gold nanoparticles function well as catalysts. This is a state that can be found in cluster structures. The Quantum size effect appears in physical attributes.

Metals with the ability to form unstable clusters in the presence of oxygen. Gold clusters, on the other hand, are extremely stable, which is why gold particles can be utilized as catalysts. Oxidation processes that are catalyzed The catalytic activity of gold nanoparticles increases as their size decreases.

Applications in energy harvesting

Because fossil fuels are nonrenewable by nature recent research has revealed their limitations and insufficiency in the coming years As a result, scientists are attempting to change their study methods. The resources that can be found readily. Produce green energy sources. Material with a low

cost Because of the following features, scientists determined that NPs were the ideal contender for these goals. a vast region of surface Photocatalytic applications is a type of catalysis that uses light. nature of catalysis Optical properties are commonly employed to generate energy through electrochemical water splitting and photoelectrochemical (PEC) reactions Other than reduction, water splitting electrochemical CO₂ and fuels, solar cells and piezoelectric generators are very advanced possibilities for producing energy NPs can store energy in many forms at the nanoscale level. As a result, they're used in energy storage applications as well. Nanogenerators have recently demonstrated the ability to transform mechanical energy into electrical energy utilizing piezoelectric technology, however, this is a novel approach to energy production.

Applications of nanoparticles in electronics

Because of the tremendous potential for lower costs than classic silicon printed processes, interest in the growth of printed electronics has been expanding in recent years.

Electronics printed with various inks will flow quickly. CNTs, organic, and ceramic nanoparticles may also be present in these inks. Instead of vacuum tubes, diodes and transistors, even tiny chips, are used to create electronic components.

Application of nanoparticles in micro-wiring

In the electronic industry, the paste of metal nanoparticles best candidate for the manufacturing of a printed wired board). The metal nanoparticles have less melting point than the bulk metals. By the conventional electric conduction paste, circuit formation is possible on polymer base material. Whenever particles are used at the nanoscale, the thickness of the wire is decreased to a nanolevel. The ink-jet method is very important for the formation of wiring at the nano level. The ink-jet method is not very expensive, and it does not require long times like conventional techniques such as photolithographic methods and vacuum evaporation that are generally used. Gold can be used to make metal nanoparticle paste. Because gold is so expensive, copper is utilized as a replacement.

Antioxidants are made from copper nanoparticles [24].

Silver nanoparticles

Silver ion or metallic silver, as well as silver nanoparticles, can be used in medicine for burn treatment, dental materials, coating stainless steel materials, textile fabrics, water treatment, sunscreen lotions, and other applications, according to the current study, and they are low in toxicity to human cells, have high thermal stability, and have low volatility.

Chemical reduction, thermal decomposition, laser ablation, and sonochemical synthesis are just a few of the methods for making silver nanoparticles that have been reported. Chemical reduction and laser ablation are the two most often used synthetic methods. The chemical reduction method involves reducing metal salts such as silver nitrate in a suitable medium using various reducing agents such as citrate, borohydride, and other reducing agents to produce colloidal suspensions with nanoparticles incorporated. Silver nitrate (90 mg) was dissolved in 500 mL distilled water and brought to a boil in Lee and Meisel's citrate reduction procedure. A 10% sodium citrate solution (10 mL) was added, and the solution was allowed to boil for 1 hour. The resultant colloid was greenish-yellow in color and had a maximum absorption rate of 420 nm.

Another approach, known as the Creightons method, uses sodium borohydride instead of citrate as the reducing agent. In this procedure, 50 mL of sodium borohydride (NaBH_4) was diluted with water from 0.00386 g of sodium borohydride (NaBH_4) in a volumetric flask. Nitrogen was bubbled through the aforesaid solution and then maintained in an ice bath to prevent degradation before continuing with the process. Then, in a 25 mL volumetric flask, 0.00442 g of silver nitrate was weighed and diluted with water to make a solution. The next step was to vigorously swirl 12 mL of the ice-cold produced aqueous NaBH_4 solution into 4 mL of the silver nitrate solution. This resulted in a faint yellow hue shift. The reaction was stirred until it reached room temperature. This method of synthesis consistently produces particles with a limited size distribution.

Silver's mode of action is linked to its interaction with thiol group molecules prevalent in bacterial respiratory enzymes. Silver attaches to the bacterial

cell wall and cell membrane, preventing the bacterium from breathing. Silver works in *E. coli* by inhibiting phosphate uptake and releasing phosphate, mannitol, succinate, proline, and glutamine from the cells [25]. Silver nanoparticles are used in Silver-coated medical devices. This could be due to the inactivation of metallic silver when it comes into contact with blood plasma, as well as the coatings' lack of endurance. The antibacterial activity of metallic silver was equally unimpressive.

Applications

Silver, in both its metallic and nanoparticle forms, is recognized to have significant antibacterial characteristics, and as a result, it has a wide range of applications.

- The Fe₃O₄ attached Ag nanoparticles can be utilized to clean water and can be quickly removed using a magnetic field to prevent environmental pollution.
- Because of its slow and steady reaction with serum and other body fluids, silver sulfadiazine promotes faster burn wound healing.
- Nanocrystalline silver dressings, creams, and gels effectively reduce bacterial infections in chronic wounds. As a wound treatment, silver nanoparticles comprising polyvinyl nano-fibers show effective antibacterial properties
- When evaluated on an animal model, silver nanoparticles were found to have improved wound healing capability, cosmetic attractiveness, and scarless healing.
- Silver-impregnated medical devices, such as surgical masks and implantable devices, have shown to be effective against bacteria.
- Antimicrobial nano paint that is not harmful to the environment can be developed.
- Preservatives made of inorganic composites are employed in a variety of products.
- Long-lasting antibacterial action of silica gel micro-spheres combined with silica thiosulfate.
- Treatment of burns and infections
- Silver nanoparticles can be used for water filtration.

- Silver zeolite is used in food preservation, disinfection, and decontamination of products.

Toxicity

Silver toxicity displays itself as argyria only when a large open wound is present and a considerable number of silver ions is used to treat it. There are no reports of silver allergy regularly. In most research, silver nanoparticles were shown to be non-toxic. However, because of their small size and changeable features, they are thought to be environmentally dangerous investigated the toxicity of various silver nanoparticle sizes on the BRL 3A rat liver cell line (ATCC, CRL-1442 immortalized rat liver cells). After a 24-hour exposure, the mitochondrial cells showed aberrant size, cellular shrinkage, and irregular shape, according to the researchers.

With increasing silver nanoparticle concentrations, the cytotoxicity of silver nanoparticles on mitochondrial activity increased.

The use of nanoparticles in biomedical and therapeutic applications has opened up a wide area for nanotechnology in fields such as electronics, engineering, medicine, and so on, according to the studies. However, the potential side effects of nanoparticles have not been well studied, so more research is needed before products related to nanomedicine are introduced to the market [26].

Albizia julibrissin

Albizia julibrissin is a tree species native to southwestern and eastern Asia, belonging to the Fabaceae family. Persian silk tree and pink siris are two frequent names for this plant. Although it is not closely related to either genus, it is also known as Lenkoran acacia or bastard tamarind. In the United States, the species is commonly referred to as "silk tree" or "mimosa," which is misleading because the former name can refer to any *Albizia* species that is most common in a given location, even though it was once included in *Mimosa* and is closely related to the *Mimoseae* family. *Albizia* is a genus of about 150 species of trees and shrubs native to Asia and Africa's tropical and subtropical areas.



Fig. 6. Persian silk tree

The leaf texture and flowers of *A. julibrissin* make it a popular ornamental plant in parks and gardens. Its leaves gradually close at night and during rainy periods, the leaflets drooping downward, earning it the modern Persian name *shabkhosb*, which means "night sleeper."

The total flavonoid content of *A. julibrissin* leaf was 35.14 mg/g. *Albizia julibrissin* is not one of the top ten herbs in terms of its use in traditional medicinal cures, but it does have some benefits that are worth mentioning.

Classification

Kingdom	Plantae
(unranked)	Angiosperms
(unranked)	Eudicots
(unranked)	Rosids
Order	Fabales
Family	Fabaceae
Subfamily	Mimosoideae
Tribe	Ingae
Genus	<i>Albizia</i>
Species	<i>julibrissin</i>

Antibacterial properties

Methanolic extracts of *A. niger* leaf samples The extraction value of julibrissin was the highest. Total flavonoid content (35.14mg/g) and flavonoid content (5.14g/100g). *A. sativa* leaf extracts were found in the majority of the *A. sativa* leaf julibrissin *P. Vulgaris* displayed the greatest inhibition zone (10.1 mm*) and was the least susceptible. *S. typhi* is the name of the bacterium. (3.5 mm*) Bacterial growth was suppressed by the stem and blossom extracts. Only at higher concentrations (MICs of 160-215 g/ml and 65-180 g/ml, respectively)

Traditional uses

Albizia julibrissin is a traditional snake bite and leprosy treatment. *Albizia* plants are used therapeutically in traditional Indian and Chinese medicine for insomnia, irritability, wounds, anti dysenteric, antiseptic, antitubercular, and other conditions.

Anti-inflammatory properties

When ethanol and hydro alcoholic extracts of *Albizia julibrissin* leaves were compared to diclofenac sodium as a control, it was discovered that the ethanol and hydro alcoholic extracts had a considerable anti-inflammatory effect. The screening strategy for obtaining the results was HRBC membrane stabilization. The suppression of membrane lysis was used as a measure of anti-inflammatory property in this study. A spectrophotometer set to 560 nm was used to determine the amount of hemoglobin in the supernatant solutions. The percentage of hemolysis was calculated by assuming the hemolysis produced in presence of distilled water as 100%. Finally, the results showed that a 1000 g/ml solution of ethanol extract of the plant inhibited membrane lysis by 60.87 percent, while standard diclofenac sodium inhibited membrane lysis by 69.56 percent at 50 g/L.

Antidepressant properties

Its primary activities include resolving depression for sedation, controlling qi for hunger stimulation, dispersing wind for improved vision, and boosting blood circulation for pain relief.

Depression, insomnia, chest tightness, indigestion and loss of appetite, wind-fire eye issues, blurred vision, back discomfort, and injuries from falls

are all common uses and indications. 3 to 9 grams in decoction, powder, or tea pills is the recommended dosage [27].

Objectives

Objectives

- Green synthesis of Silver Nanoparticles (AgNPs) by using *Albizia Julibrissin* aqueous leaf extract.
- Characterization of Synthesized AgNPs by UV-vis spectroscopy, DLS, Zeta Potential, FTIR, and TEM.
- Competitive Antibacterial analysis of *Albizia Julibrissin* mediated AgNPs and *Albizia Julibrissin* aqueous leaf extract.

Materials and Methods

Materials and methods

All chemicals used in this experiment were of the highest purity and obtained from Sigma(Bangalore, India) and Merck(Mumbai, India). leaves were harvested from the campus of Integral University Lucknow, India for silver nanoparticle synthesis.

Plant extract and synthesis of silver nanoparticles

For the preparation of *Albizia julibrissin leaf* extract, leaves were thoroughly washed and dried completely and weighed about 5gm then leaves were crushed in a pestle mortar and 20 ml of distilled water was added to it and all leaves were crushed properly. Take some ice cubes in the polypropylene molded tray and place pestle & mortar in it with plant extract and leave it for some time. Then again crush the extract and filter it with the help of Whatman filter paper in the centrifuge tube and then the tubes were placed in the centrifuge at 6000 rpm at 4°C for 10 min. Then remove the pellet from the extract and take the supernatant into another centrifuge tube. The extract is stored in a refrigerator for future purposes.

3ml of the prepared plant extract was taken in 20 ml of the centrifuge tube and 3 μ l of silver (stock 1mg/ml) was added to the plant extract. Keep the reaction tube in the incubator at about 37°C for 48 hrs.

After 48 hrs, plant extract changes its color due to silver salt from yellow to gold indicating the synthesis of silver nanoparticles. Then the sample was taken for UV/VIS spectroscopy and O.D values were recorded, sample was stored in the refrigerator for further experiments.

Results

Result

The alkalinized extract of *Albizia julibrissin* possesses a broad variability of metabolites that may help in the reduction of silver nitrate. The preliminary confirmation for the formation of AgNPs was the visual observation of the color change of the reaction mixture. The color, noted by visual observation, increased in intensity resulting in a brownish-yellow color from the original yellow after 24h of incubation (figure 1). The color change was noticed a few minutes after the addition of AgNO₃, but the intensity increased with time. Similar color changes have been observed in previous studies thereby confirming the completion of the reaction between the alkalinized extract and AgNO₃. Characterization of AgNPs using various techniques such as UV-Vis Spectroscopy, Dynamic Light Scattering, Zeta Potential, and Transmission Electron Microscopy.

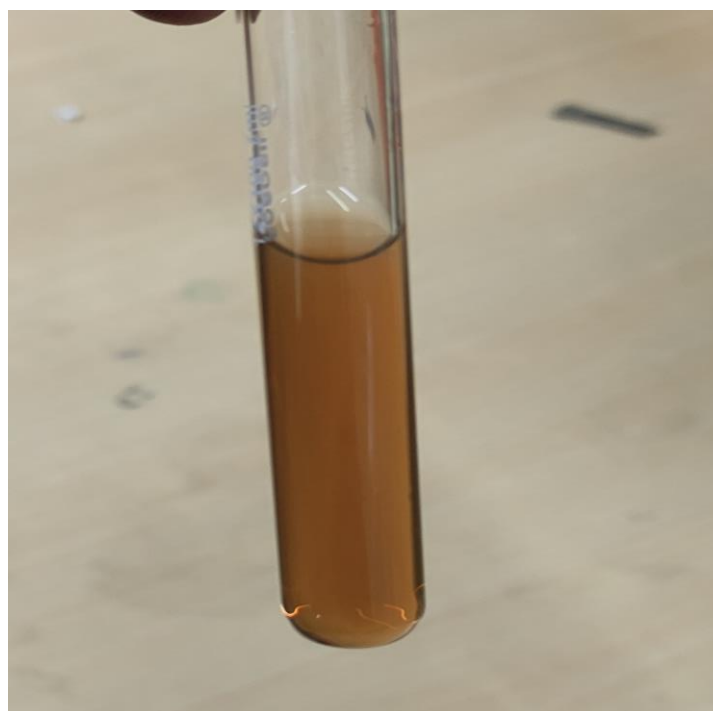


Fig. 7. Picture shows the reduction of silver nanoparticles (brown color).

Uv/vis spectroscopy

UV/Vis spectrophotometric measurements were performed on a Shimadzu dual-beam spectrophotometer operated at a resolution of 1nm in the quartz cuvette. Ultraviolet-visible spectroscopy or ultraviolet-visible spectrophotometry (UV-Vis or UV/Vis) refers to absorption spectroscopy or

reflectance spectroscopy in the ultraviolet-visible spectral region. The absorption or reflectance in the visible range directly affects the received color of the chemicals involved. In this region of the electromagnetic spectrum, atoms and molecules undergo electronic transitions. Absorption spectroscopy is complementary to fluorescence and deals with transitions from an excited state to the ground state, while absorption measures transitions from the ground state to the excited state.

To observe the optical property of biosynthesized silver nanoparticles, samples were periodically analyzed for UV- vis spectroscopic studies at room temperature operated at a resolution of 1nm between 250 and 800nm ranges.

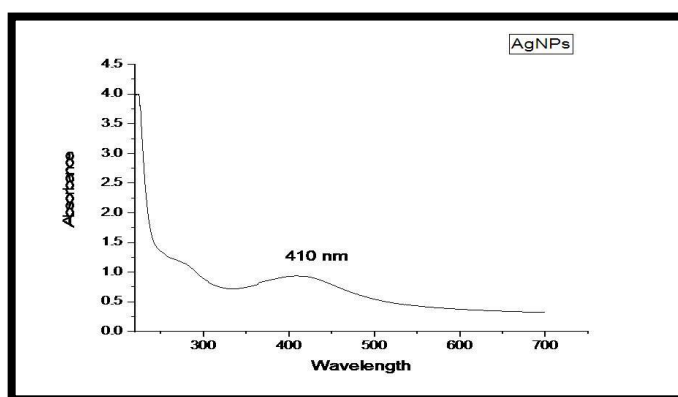


Fig. 8. UV-Visible Spectroscopy graph of AgNPs shows an absorption band at 410 nm.

The synthesis of silver nanoparticles by the reduction of aqueous metal ions during the exposure of alkalinized *Albizia julibrissin* leaf extract was easily monitored by using UV-Vis spectrophotometry. In this study, the formation of AgNPs was monitored by measuring the UV-Vis spectra at different time intervals (figure 2). As the time increased, the intensity of the absorbance increased, indicating an increase in the amount of AgNPs produced by the mixture. A UV absorption spectrometric analysis of the AgNPs showed absorbance spectra at 410 nm, suggesting the bioreduction of silver nitrate into silver nanoparticles.

DLS (Dynamic Light Scattering)

The thin electric dipole layer of the solvent adheres to the surface of a dispersed nanoparticle when it moves through a liquid medium; therefore,

the hydrodynamic diameter estimated by DLS provides us information about the inorganic core along with coating material and the solvent layer attached to the particle.

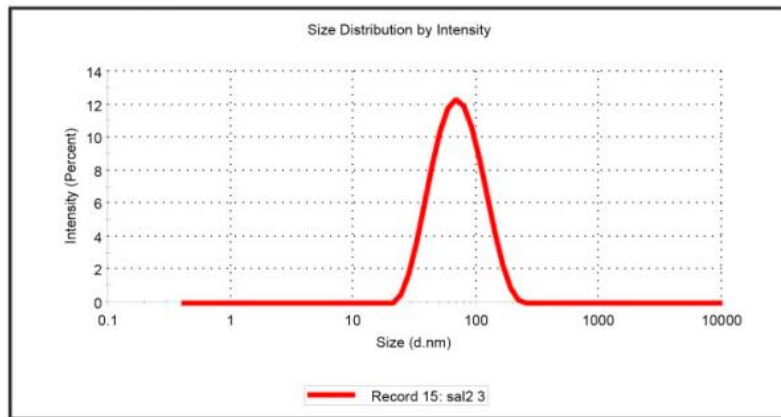


Fig. 9. Size distribution of colloidal AgNPs. (average size 25d.nm).

Zeta Potential

The determination of Zeta Potential is considered an effective, simplest, and most straightforward method to predict the stability and understand the surface properties of the nanoparticles. Information regarding the concentration, distribution, exposure, or shielding of charged moieties, ionization, and adsorption could be drawn from the analysis of zeta potential.

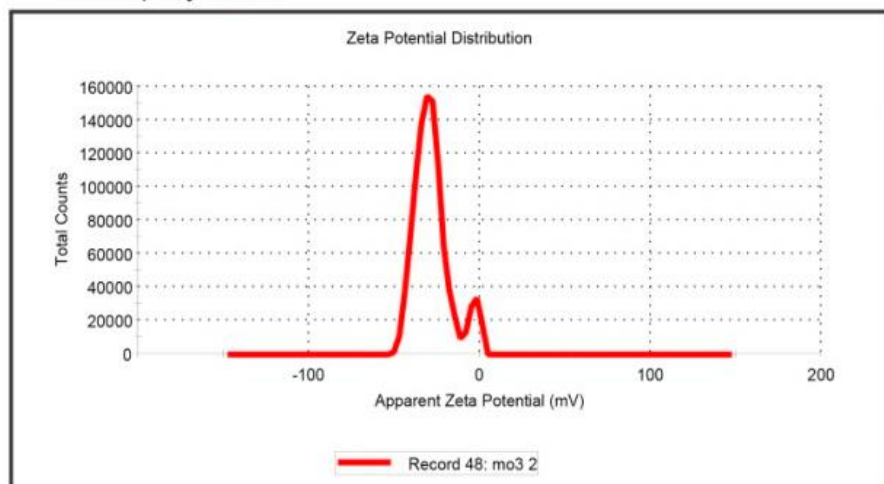


Fig. 10. Zeta potential graph shows the value of AgNPs at -25mV.

Transmission electron microscopy

The high-resolution image was acquired using the transmission electron microscope (TEM), which confirmed the average size of AgNPs as 29 ± 2 nm using a Gatan digital micrograph and showed the spherical form of AgNPs.

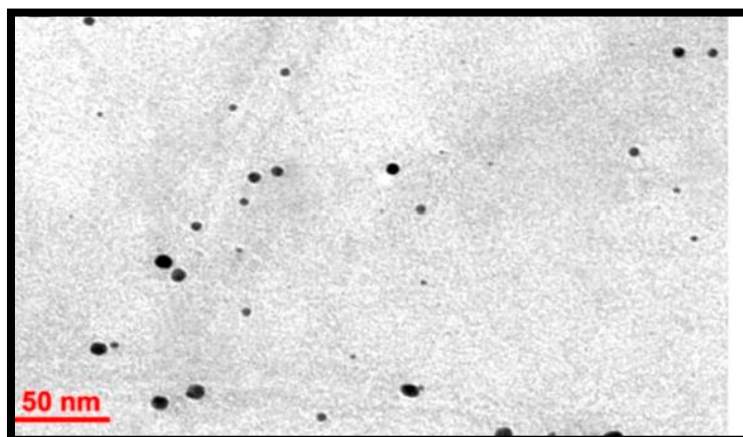


Fig. 11. TEM analysis of synthesized nanoparticles (29 nm).

Antibacterial screening

The antibacterial action of synthesized AgNPs against both gram-positive and gram-negative bacterial strains was found to be satisfactory. Using the agar well diffusion method, the antibacterial potential of synthesized AgNPs was evaluated against normal strains of *Pseudomonas aeruginosa* and *Staphylococcus aureus*. The antibacterial potential was confirmed by a clear zone of inhibition surrounding the inoculated region.

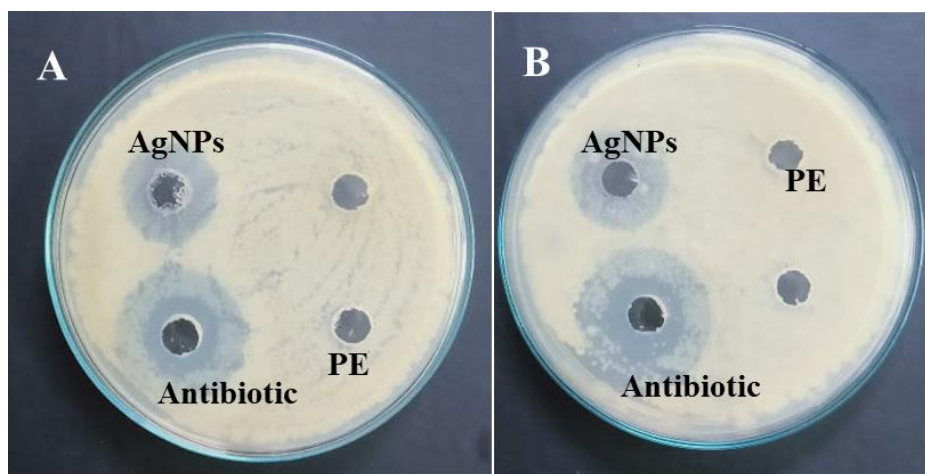


Fig. 12. Synthesized AgNPs shows Antibacterial activity against (A) *Pseudomonas aeruginosa* and (B) *Staphylococcus aureus*.

S.No.	Bacterial strain	ZOI (mm)	
		AgNPs	Antibiotic (levofloxacin)
1.	<i>Staphylococcus aureus</i>	20	31
2.	<i>Pseudomonas aeruginosa</i>	25	30

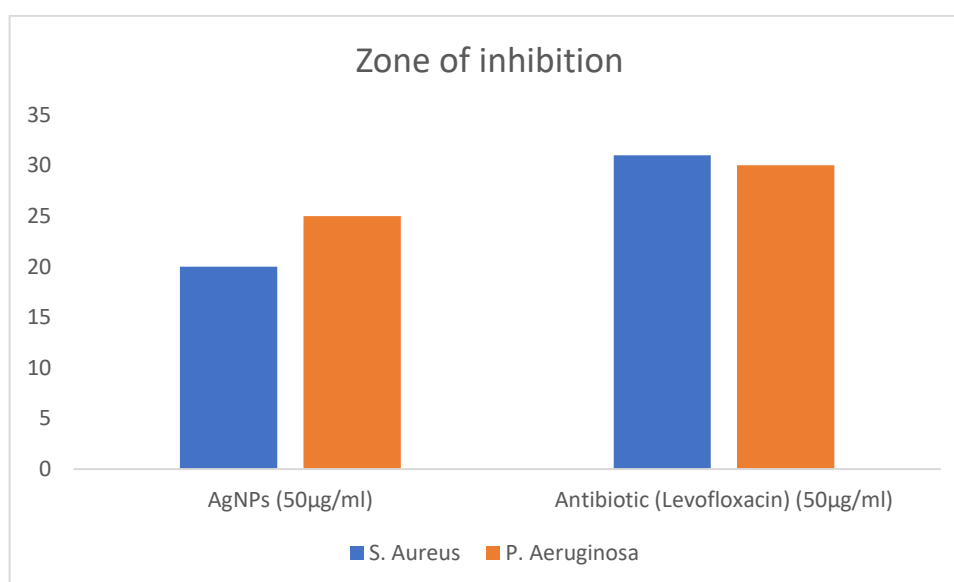


Fig. 13. Table and graph showing zone of inhibition of AgNPs against the above-mentioned pathogens..

MIC (Minimum Inhibitory Concentration)

We found that biogenic AgNPs synthesized by *Albizia julibrissin* leaf extract show strong antibacterial activity against both Gram-positive and Gram-negative pathogenic bacterial strains. The MIC₅₀ of AgNPs was evaluated against different pathogenic bacterial strains that included 1.9 µg/mL against *S. aureus*, and 8.6 µg/mL against *E. coli*, indicating its broad-spectrum feature. However, we found that AgNPs were more effective against *E. coli* (Gram-negative) and *S. aureus*, (Gram-positive) than other pathogenic strains. A thick peptidoglycan layer in Gram-positive bacteria prohibited the entry of AgNPs into the cytoplasm, and a higher AgNPs

concentration is required to inhibit the growth of Gram-positive than Gram-negative bacteria.

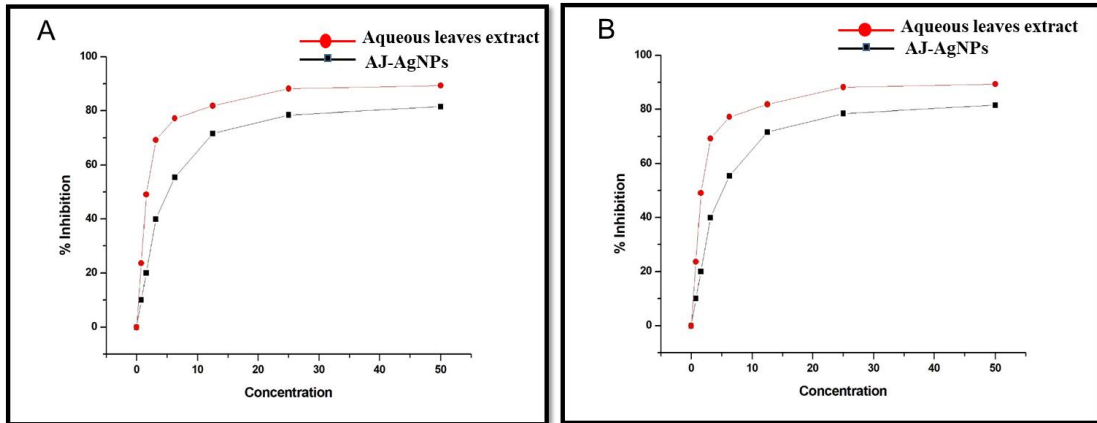


Fig. 14. IC₅₀ (8 µg/ml) of Antibacterial potential of AgNPs against (A) *Staphylococcus aureus*, and (B) *Pseudomonas aeruginosa* (96 well plate method).

Discussion and conclusion

Discussion and conclusion

Albizia julibrissin leaf extract was utilized as a reducing and stabilizing agent in this work. The synthesis of AgNPs is thought to be triggered by *Albizia julibrissin* aqueous leaf extract's reducing enzymes and capping agents, such as secondary metabolites, which work together to decrease AgNO₃. The color of the synthesized AgNPs was the brown color which is the characteristic feature of AgNPs. According to Mie Theory, silver shows resonance known as Plasmons in UV- the visible spectrum. These resonances are formed by the interaction of electromagnetic waves and electrons at the surface of AgNPs. This resonance characteristic of AgNPs can be observed by spectroscopy.

The antibacterial activity of AgNPs was determined against four species of Gram-negative e pathogens: *Pseudomonas aeruginosa* and *Staphylococcus aureus*. The results of the disk diffusion test, of the AgNPs are summarized in Table. For the disk diffusion test, the presence of a clear zone around the AgNPs disk suggests that the AgNPs possessed antibacterial activity which can inhibit the growth of the Gram-negative foodborne pathogens. The visible clear zone produced by AgNPs against four different species of Gram-negative and gram-positive bacteria is shown in Figure.

Conclusion

A simple, one-step green approach was developed for the synthesis of AgNPs using, *Albizia julibrissin* leaf extracts the plant extract acts as both a reducing and stabilizing agent.

Silver nanoparticles showed significant antibacterial activity against the selected Gram-negative and gram-positive pathogens. Thus, AgNPs might be a good alternative to develop as an antibacterial agent against the multidrug-resistant strains of bacteria. The applications of AgNPs may lead to valuable findings in various fields such as medical devices and antimicrobial systems.

A critical need in the field of nanotechnology is the development of a reliable and eco-friendly process for the synthesis of metallic nanoparticles. Nanoparticles are being viewed as fundamental building

blocks of nanotechnology. Silver nanoparticles play a profound role in the field of biology and medicine due to their attractive physicochemical properties. The silver nanoparticles synthesized from alkalized *Albizia julibrissin* leaf extract by a bio-reduction method exhibit all the characteristic features of nanoparticles.

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