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GREEN SYNTHESIS OF NANOPARTICLES: A BRIEF STUDY

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ABSTRACT: Nanoparticles are small particles ranging from 1 to 100 nanometers. The biological production of nanoparticles is cheaper than chemical production. Besides being cheaper, it is eco-friendly as well. This biological procedure is a bottom-up procedure where the extracts of natural products replace the chemical reducing agents. These reducing and stabilizing agents are found in plants and can be used to synthesize nanoparticles. Plants also have growth terminating and capping properties as well. This paper aims to give brief information on different plant sources and synthesis of nanoparticles. The infection caused by multi-drug-resistant bacteria increases the mortality rate. So, the effect of multi-drug-resistant bacteria is gaining much more importance. The nanoparticles synthesized by green sources are non-toxic to the environment and humans. This review focuses on basic knowledge of nanoparticles, their production by green sources, the application against multi-drug-resistant bacteria, and advantages and disadvantages of nanoparticles.

INTRODUCTION: Nanotechnology is currently a multidisciplinary field encompassing engineering, biology, chemistry, and physics [1, 2]. Nanotechnology deals with the processing of separation, consolidation, and deformation of material by one atom or by one molecule "was well defined by Professor Norio Taniguchi" [3]. It uses nanoscale particles and materials for use in different spheres of life. It deals with governing matter at the molecular level and has firmly entered the realm of the vast area of application. Nanotechnology is playing a critical role in many significant technologies *via* nanoscale structures in areas of optics, electronics, biomedical sciences, mechanics, drug-gene delivery, chemical industry, optoelectronics devices, non-linear optical devices, catalysis, space industries, energy sciences, and photo-electrochemical applications [4]. It has also been used in cosmetics, packaging, drugs, *etc*. Nanoparticles are ultrafine particles that range from 1-100 nanometers (nm) in diameter. They possess remarkable and interesting properties owing to small sizes, large surface area with free dangling bonds, and higher reactivity over their bulk cousins [5]. For years, nanoparticles have been produced from chemicals, but nowadays, plant extracts have replaced these chemicals.

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The effects of the nanoparticles on the biological methods are also considered because all the biological processes are carried out at the nanoscale range. Therefore these particles are compatible to work alongside, and secondly, the nanoparticles are effective against the prokaryotes due to their physical and chemical properties. Due to those precise characteristics, they're capable of counteracting the microbes without problems; however, they are not yet proven to be harmful to model organisms when used in an appropriate dose. In recent years, nanoparticles' natural extract synthesis approach has become a popular methodology because of their various antimicrobial characteristics [6]. Chemical, physical and biological (natural precursors) have been analytically studied for the design and synthesis of nanoparticles with the desired size, shape, and functionalities [7, 8, 9, 10]. The use of green sources has two advantages over chemical methods. Firstly, these are much more economical than chemical reagents, and secondly, these plant extracts would not harm the body. The green synthesis of nanoparticles is successful because the plant extract contains biomolecules like protein, amino acids, sugars, water soluble carbohydrates, terpenoids, and flavonoids, which play the dual role of reducing the chemicals to form nanoparticles and stabilizing it [11]. Plant extract-based nanoparticle synthesis uses various natural products which can produce nanoparticles with more purity [12]. These techniques are cost-effective, and eco-friendly and avoid complex procedures [13, 14, 15, 16, 17, 18]. UV-vis spectroscopy is done after the synthesis to validate the synthesis of nanoparticles.

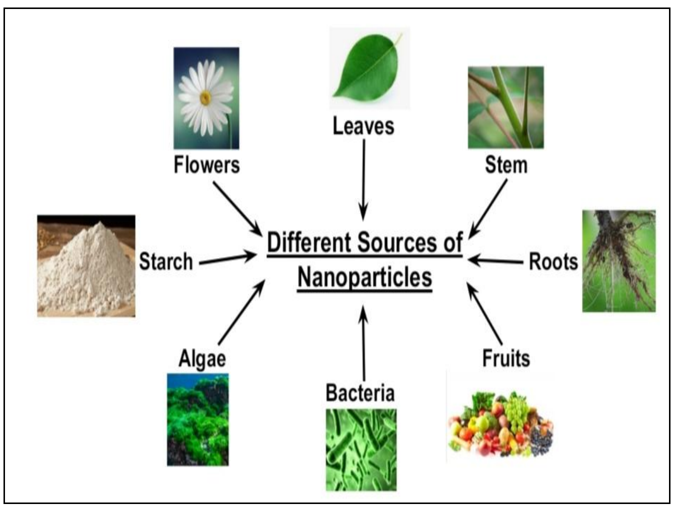

FIG. 1: DIFFERENT SOURCES OF GREEN SYNTHESIS OF NANOPARTICLES

Nano-Science: It has been years since plants have been used to reduce metal ions. Nanotechnology is a branch of science that deals with nanoparticles, their properties, their formulation, and their use, especially in the medical field. Nanoparticles are colloidal-sized particles composed of polymers with diameters ranging from 10 to 1000 nm[19]. They have multifunctional properties, so they can be used in different fields. They are grouped into solid lipid nanoparticles, polymeric nanoparticles, ceramic nanoparticles, hydrogel nanoparticles, co-polymerized peptide nanoparticles, nanocrystals and nanosuspensions, nanotubes and nanowires, functionalized nanocarriers, nanospheres and nanocapsules [20]. The nanoparticle delivery systems improve skin penetration, release and surface functionalization of active compounds. They may contain metallic ions. Metallic nanoparticles containing iron oxide, gold, silver, gadolinium, and nickel are used to formulate cosmetics, sunscreens, and personal care products [20]. Nano-medicines have a large impact on the treatment of various diseases. Hence, the synthesis of nanoparticles is considered the building block for future generations in combating various diseases.

Green Synthesis: The three most important conditions for the synthesis of nanoparticles are:-

- Eco-friendly solvent.
- Good reductant.
- Non-toxic material for stabilization.

Green synthesis is a promising method for nanoparticle synthesis because of the utilization of cost-effective and non-hazardous raw materials [21]. This nanoparticle synthesis method is sustainable with great therapeutic efficacy, reduced toxicity, targeted binding, and site-specific delivery [22]. The biological route is safe as well. This does not affect human health and the environment [23, 24]. This green synthesis of Nanoparticles can be obtained from various biological sources such as stems, flowers, fruits, leaves, fungi, bacteria, seeds, roots, etc.

Moreover, in most cases, a reducing agent from the plant extract plays a role as the capping and stabilizing agents [25, 26, 27]. The researchers have reported that the size and morphology of nanoparticles can be altered by changing the synthesis parameters, such as metal salt, pH, temperature, and reaction time [28]. It is a cost-effective, rapid, environmentally benign, and biocompatible process, thus safe for clinical research [22]. It is a promising route for the production of biocompatible, stable nanoparticles. Green nanoparticle synthesis has also employed amino acids, phytochemicals, polysaccharides, polyphenols and vitamins [29, 30, 31]. These nanoparticles have great potential for target drug delivery and can exhibit antiviral, antibacterial, and anti-cancer properties [32, 33, 34, 35, 36]. Compared with materials that have an undefined particle size, Nanomaterials consist of small particles with a large specific surface area, resulting in materials that display unexpected surface area, volume, quantum size, and macro tunneling effects [37]. Nanomaterials exhibit unique optical, mechanical, catalytic, and biological properties because of these characteristics, resulting in nanomaterials having broad application potential [38]. Plants and their extracts are uncomplicated to obtain, and the technique calls for the handiest salt solution is the answer as a metal precursor. In general, metal nanoparticles, particularly Gold Nanoparticles, have been used in various applications, such as drug delivery, molecular imaging, and cancer diagnostics and therapy, which depend on the exploitation of important sources, mainly microbes, plants, and fruit waste [39]. Currently, interest is focused on the antiviral activity of silver Nanoparticles given the COVID-19 pandemic [40, 41]. Proteins are also used as a bio-reducing agent for metallic ions, which have many advantages and disadvantages. They have been effective in controlling the shape and size of nanoparticles [42]. Utilizing water as a solvent diminishes the noxious effect of chemical solvents [43]. The drawbacks of using these residences include excessive protein sensitivity, their changing configuration, and reduced performance. Proteins are also temperature sensitive, showing resistance at high temperatures [44].

To avoid further toxicity of surfactants that act as the shape controlling agents, the bio-inspired synthesis of nanoparticles can be carried out instead [45]. Different physical and biochemical approaches are reported to produce individual or hybrid nanoparticles utilizing plant extracts [46, 47, 48, 49, 50]. There are special methods for the synthesis of nanoparticles. They are as follows:-

Bottom-Up approach: In the bottom-up approach, usually chemical and biological routes are used to synthesize nanoparticles by self-assembly of atoms to initiate new nuclei, which grow into nanosized particles [51, 52, 53]. In this process, nanoparticles are processed from small units such as atoms or molecules or the self-assembly of atoms to form new nuclei with nanoscopic dimension.

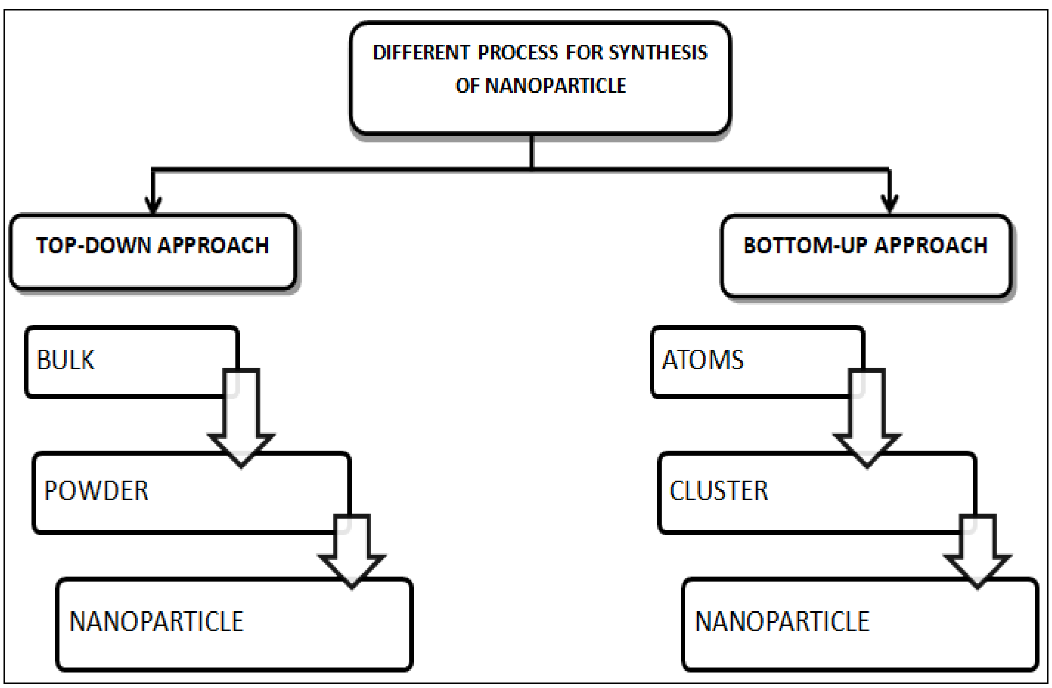

FIG. 2: DIFFERENT APPROACHES TO SYNTHESIS OF NANOPARTICLE

Top-Down Approach: In top-down methods, sample bulk material is broken down into smaller fine particles by size reduction using different approaches such as ion and ball milling, /laser ablation, *etc.* [51, 52, 53]. This process reduces bulk materials to form small units by various lithographic methods like crushing, spitting, and milling.

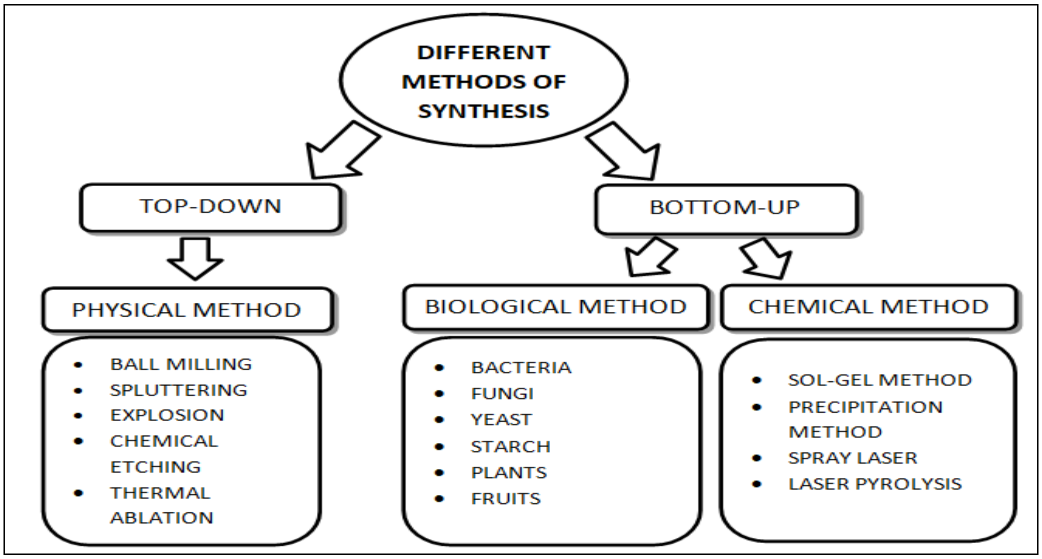

FIG. 3: DIFFERENT METHODS OF SYNTHESIS OF NANOPARTICLE

The stability, shape, and size of nanoparticles can be precise by controlling the temperature, pH, and concentration of plant extract and meat salt solution as well as incubation [1].

Tables 1 and **2** summarize different types of nanoparticles obtained by different green sources such as plant, bacteria, etc., along with the size of the nanoparticles.

TABLE 1: DIFFERENT TYPES OF NANOPARTICLES OBTAINED FROM VARIOUS PLANT SOURCES

Sl. no.	Nanoparticle	Plant Source	Size
1	Copper And Its Oxides	*Citrofortunellamicrocarpa* Leaf Extract[54]	54-68nm (Spherical)
2	Copper And Its Oxides	*Pterolobiumhexapetalum* Leaf Extract [55]	10-50nm (Spherical)
3	Copper And Its Oxides	Flower Extract of *Euphorbia pulcherrima*[56]	19.2nm (Cubic)
4	Copper And Its Oxides	*Rosa canina* fruit extract[57]	15-25nm (Spherical)
5	Copper And Its Oxides	*Hylotelephium telephium* Flower Extract[58]	83nm (Spherical)
6	Copper And Its Oxides	Leaf Extract of *Terminaliacatappa L.*[59]	29-103nm (Spherical)
7	Cerium Oxide	*Ceratonia siliqua*[60]	22nm (Spherical)
8	Cerium Oxide	*Morus nigra* Fruit Extract [61]	~7.5nm (Irregular)
9	Iron Oxide	*Sida cordifolia*[62]	15-18nm (Spherical)
10	Iron Oxide	*Ruelliatuberosa*[63]	52.78nm
11	Iron Oxide	*Malvasylvestris*[64]	-
12	Iron Oxide	*Citrus sinensis* [65]	-
13	Iron Oxide	Monocotyledenous Plant Hordeum[66]	<30nm (unstable)
14	Iron Oxide	Dicotyledenous Plant Rumex[66]	10-40nm (highly stable)
15	Cadmium Oxide	Turmeric Plant Extacrt [67]	38.36-77.91nm
16	Titanium Dioxide	Leaf Extract of *Ecliptaprostate*[68]	36-38nm
17	Silver	Leaf Extract of *Allium paradoxum*[69]	5nm (30 min)
18	Silver	*Fritillaria* Flower Extract[70]	5-10nm
19	Silver	*Mentha pulegium*[71]	1nm (10 min)
20	Silver	Orange blossom extract[72]	5-40nm
21	Silver	Waste Extract of *Carthamus tinctorius L.*[73]	8.67 ± 4.7 nm (Spherical)
22	Silver	Leaf Extract of *Hippophae rhamnoides*[74]	2.5-14nm
23	Silver	Leaf Extract of *Ctenolepisgarcini*[75]	20-50nm (Cubic)
24	Palladium	Urtica[76]	7.44 ± 1.94 nm
25	Palladium	*Padinaboryan*[77]	8.7 nm (Crystal)
26	Palladium	*Fritillariaimperialis*flower Extract[78]	10-20nm
27	Palladium	Root Extract of *Euphorbia candylocarpa*[74]	-
28	Ruthenium	Leaf Extract of *Gloriosasuperba*[79]	36nm
29	Zinc Oxide	*Arthrospiraplatensis*[80]	≈30.0 to 55.0 nm
30	Zinc Oxide	*Elaeagnus angustifolia L.* Leaf Extract[81]	~ 26 nm
31	Zinc Oxide	*Phoenix dactylifera*[82]	30 nm
32	Zinc Oxide	*Brassica oleracea* [83]	70^{0}C
33	Zinc Oxide	*Rhamnus virgate*[84]	-

TABLE 2: DIFFERENT TYPES OF NANOPARTICLES OBTAINED FROM VARIOUS GREEN SOURCES

Sl. no.	Nanoparticle	Source	Size
1	Zinc Oxide	*Streptomyces sp.*[85]	-
2	Cerium Oxide	Fungal mediated[86]	12-20nm
3	Cerium Oxide	Agarose[87]	10nm
4	Cadmium	Fungus *Coriolus versicolor*[88]	100-200nm
5	Cadmium	*Lactobacillis sp*[89]	4.93nm
6	Cadmium	*Saccharomyces cervisiae*[89]	3.75nm
7	Cadmium	*Rhodobacter sphaeroides*[90]	2.3nm,6.8nm,36.8nm
8	Silver	Honey[91]	-
9	Silver	Rhizome extract of *Dioscorea batatas*[92]	-
10	Lead Sulfide	Bacterial strain Ns2 and NS6[93]	-
11	Lead Sulfide	Methanolic extract of *Coccus nucifera*[94]	47nm

Importance of Plants in Synthesis of Nanoparticles: In the biosynthesis of nanoparticles environmentally accepted "inexperienced chemistry" idea has been implemented for the improvement of clean and surroundings pleasant nanoparticles, which include bacteria, fungi, plants, actinomycetes, *etc*. that's stated to be "Green Synthesis" [95]. The biosynthesis of nanoparticles by using different organisms epitomizes a green substitute for the invention of nanoparticles with innovative properties. Plants have revealed outstanding potential in heavy metal detoxification and accumulation by which environmental pollutants problem can be overcome because very small traces of these heavy metals are also toxic even at low concentrations [96].

The use of nanoparticles in cancer treatment has substantially increased drug delivery to the target compared to conventional drug administration methods [22]. Effective targeting, delayed release, prolonged half-life, and reduced toxicity are the significant benefits of nanomedicine and delivery systems [97]. One advantage of using plant as a nanoparticle source is that the kinetics is higher than that of any other chemical-mediated preparations of nanoparticles. They are also non-toxic and compatible with skin and therefore are suitable for use as additives of products designed to be in contact with the human body [37]. Antioxidants in plants are reductive, consisting of polysaccharides, polyphenols, flavonoids, vitamins, amino acids, alkaloids, tannins, saponins, and terpenoids. Compared with physical and chemical methods, the preparation of Nanoparticles from plant extracts can be controlled so that NPs obtained have a specific size and morphology [98]. Other factors also affect the morphology of Nanoparticles: the concentration of plant extract and precursors, duration of reaction, pH value, and calcination temperature [37]. Many nanoparticles such as gold, silver, zinc oxide, and iron have been synthesized very easily by adopting a green approach [99].

Properties of Nanoparticles: The properties of the nanoparticles can be characterized by several methods. These methods include SEM, TEM, XRD, EDX, FTIR, UV Vis Spectroscopy, and many more. The SEM technique is based on the electron scanning principle and provides all available information about the nanoparticle at nanoscale level [100]. An electron microscope produces images by scanning the sample's surface with a beam of electrons. The scanning electron microscopy (SEM) analysis showed that the nanoparticles agglomerated, forming spherical-shaped particles. Similarly, TEM is based on the electron transmittance principle to provide information of the bulk material from very low to higher magnification [100].

Transmission electron microscopy (TEM) is broadly used to examine the scale of small nanoparticles *via* imaging, revealing phase/ crystallographic orientation records *via* a diffraction sample and coming across chemical composition through the strength spectrum. The size and morphology are measured through a transmission electron microscope (TEM) operated at an accelerating voltage of 115KV. So far, AFM has been proven to be a powerful tool to measure the adhesion and friction between a nanoparticle and a solid surface. UV-vis spectroscopy is done to measure its optical density at 440 nm and the peak is obtained at 200-900nm. FTIR measurement is recorded to obtain the chemical group present around the nanoparticle for stabilization.

XRD is also used for the characterization of the nanoparticle, which is an important characterization technique to explain the structural properties of Nanoparticles. It gives enough information about the crystallinity and phase of Nanoparticles [100].

Different Methods for the Production of Nanoparticles:

Polysaccharide Method: For the synthesis of nanoparticles, three aspects are required:

- Solvent
- Reducing agent
- Capping agent

In some cases, polysaccharides act as both reducing and capping agents. Chitosan is a polysaccharide obtained by partial deacetylation of chitin. Heparin is extensively medical used as an anticoagulant because of its precise chemical structure. Polysaccharides are a sizable magnificence of hydrophilic polymers with herbal beginning and biocompatibility that discover common use in water-primarily based polymer structures. In nanotechnology, that is particularly because of their beneficial residences in organic structures, *e.g.*, biodegradability, biocompatibility and low toxicity [101]. To correctly integrate the blessings of polysaccharide NPs and polymers, graft modification/copolymerization techniques are necessary. Two sorts of grafting reactions may be carried out to shape the polymer-grafted polysaccharide system, *i.e.*, the "grafting to" and "grafting from" reactions [102].

Tollen's Method: Tollen's synthesis method uses Tollen's reagent $[Ag(NH_3)2]+$ as a source of silver ion and aldehyde as a reducing agent. This method produces controlled-sized silver nanoparticles in a single-step process. In this green synthesis technique, the size and morphology of Silver Nanoparticles were controlled by changing the concentration of ammonia and the nature of the reducing agent [103].

Irradiation Method: Silver nanoparticles can be synthesized by using the irradiation method. The interaction between irradiation energy and the irradiated solution has been the key to controlling the properties of Nanoparticles by kinetics reaction manipulation. However, high energy is needed to ionize those systems; additionally, a complicated experimental setup is required, making manufacturing non-rentable for industrial production [104]. However, ultrasonic irradiation power is the main factor of chemical intermediates generation at pressure-temperature relationships during the oxidation-reduction reaction [105].

Biological Method: This method uses extracts from bio-organisms to synthesize Nanoparticles. This bio-organism extract acts as both a capping and reducing agent. This biologically mediated nanoparticle synthesis can be done using plants, bacteria, fungi, *etc*. The extracellular synthesis of the nanoparticles/crystals in the culture solution is called biologically induced biomineralization [106]. Suppose the synthesis happens within the intracellular space of a magnetotactic bacteria or sulfur-reducing bacteria. In that case, the process is called biologically controlled biomineralization (BCM), and the synthesis happens at a specific site within the cytoplasm or the cell wall [106].

Polyoxometalates Method: In this method, Polyoxometalates act as both a reducing agent and a stabilizer. Polyoxometalates (POMs) are good end-capping ligands for gold nanoparticles [107]. The POM's size, charge and functionality control the resulting POM–AuNP hybrid structure[107].

Advantages of Nanoparticles:

- These synthetic pathways do not use toxic solvents, chemical precursors, and additional reducing agents [108, 109].
- This plant-mediated synthesis of nanoparticles is toxic-free, environment-friendly, and has the potential to produce on a large scale and cost-effective.
- It has the advantages of environmental sustainability [37].
- Nanoparticles (NPs) have significant advantages such as small particle size, good stability, improving the insoluble drug's solubility, and reduced drug toxicity [110, 111].
- These green approaches have enough potential to develop well-defined nanoparticles of a particular size and definite shape for various

promising potential applications at commercial level [42].

- The combination of nanotechnology and green chemistry has extended the various range of biologically compatible metallic nanoparticles [42].
- They increased the therapeutic efficiency as well as bioavailability [112].

Disadvantages of Nanoparticles:

1. Nanoparticles increase health issues as inhalation of nanoparticles can cause severe lung diseases.
2. They also increase pollution which is known as "Nano pollution," which is very dangerous to living organisms.
3. The development of nanotechnology leads to the loss of jobs in the traditional farming and manufacturing industry.
4. Atomic weapons can now be more accessible and made more powerful and destructive [112].
5. Producing nanoparticles for drug delivery, the use of extensive polyvinyl alcohol as a detergent can create an issue of toxicity [112].
6. The production of nanoparticles at large is very expensive, which is why the products made up of nanotechnology is more expensive.

Antimicrobial Property of Nanoparticles: Bacteria are small, single-celled micro-organisms ubiquitous in nature. Pathogenicity is the ability of bacteria to cause diseases. Some bacteria are useful to the human body, but some can be fatal. Nanoparticles with antimicrobial activity that combats *Enterococcus facium, Streptococcus aureus, Klebsiella pneumonia, Acinetobacter baumannii, Pseudomonas aeroginosa, Enterobacter sp.* include nanoparticles containing Silver, Gold, Zinc, Nickel, Cerium, Selenium, Aluminium, cadmium, Yttrium, Palladium or super magnetic Iron [113]. Silver nanoparticles are used to a great extent since they have multiple mechanisms of antibacterial actions, high compatibility, and functionalized potential and are easy to detect [114]. Increasing antibiotic resistance causes bacteria to seriously threaten human life [115]. The green synthesis of ZnO NPs using plant extracts would overcome the problem of antibiotic resistance related to the formation of bacterial biofilms [116], and because of their good antibacterial properties and biocompatibility, non-toxic, safe, and stable characteristics they will become a new research direction in the field of antibacterial agents. The effect of nanoparticles on the cells of living organisms depends on their diameter, size, and shape [117]. Larger nanoparticles are more toxic due to their easier absorption and larger surface area. Furthermore, the metal ion's release rate increases as the size of nanoparticles decrease [118]. Hence it can be said that the toxicity of the nanoparticle depends on its size.

Application of Nanoparticles: Nanoparticles possess antimicrobial activity that can overcome common resistant mechanisms, including enzyme inactivation, decreased cell permeability, modification of target sites/enzymes and increased efflux through overexpression of efflux pumps, to escape from the antibacterial activity of antimicrobial agents [119]. Moreover, nanoparticles conjugated with antibiotics show synergistic effects against bacteria, prohibit biofilm formation and have been utilized to combat MDROs [119]. For the protection of public health, antibacterial materials are increasingly demanded in various applications, such as sportswear and medical and protective textiles to prevent bacterial infection [120].

NPs can be incorporated as antibacterial agents in green, non-toxic, and harmless antibacterial textiles and are effective in reducing cross-infection, preventing the spread of disease, and thereby ensuring the safety and health of humans [37]. However, the complexity of the physio-chemical properties, including size, shape, chemical modification, solvent, and environmental factors, can affect the antibacterial properties of nanoparticles during the preparation of nanoparticles and interaction with bacteriananoparticles coating an implantable device, wound dressings, bone cement, or dental materials can function as nanoparticle-based antibiotics delivery system [121].

CONCLUSION: The production of nanoparticles from green sources is increasingly important. In recent studies, the main focus is in the synthesis of nanoparticles from green sources, which is easier and cheaper than chemical sources. The production of nanoparticles from green sources is eco-friendly, inexpensive, and free of chemical contaminants.

Furthermore, they have a wide area of applications such as the medical field, optics, catalysis, sensors and many more. With evolving technology, the use of nanoparticles will be used in the treatment of many diseases and will replace the traditional antibiotics in the future. These can be used in replacement of traditional antibiotics because it has anti-microbial property, which will not allow micro-organism growth. Nanoparticles itself has so many benefits. These biological methods are non-toxic to the environment as well as to humans. Moreover, nanoparticles synthesized by green sources are more stable and effective. With controlled experiments, nanoparticles of derived size can easily be obtained. The large-scale production of nanoparticles can fulfill the future demands of a growing population. Metal nanoparticles produced by plants and/or plant extracts are more stable as compared to those produced through different organisms.

Plants contain some unique compound that not only helps synthesize nanoparticles but also increases the synthesis of nanoparticles. Plants extracts can reduce metal ions faster than plants, fungi, or bacteria. Nowadays, several nanoparticles other than silver are being synthesized, such as Iron, Cadmium, Titanium, Copper, Cerium, zinc, etc. These nanoparticles have imperative roles in human welfare. This review gives information about nanotechnology, which deals with nanoparticles, their synthesis from different green sources, and their advantages, disadvantages, and application.

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