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A Review of Respiratory Adaptations in Aquatic and Terrestrial Chordates

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Abstract
Green synthesis of metal nanoparticles (MNPs) using microbial and phyto-extracts has emerged as a sustainable and biocompatible approach for developing advanced nanomaterials with significant biomedical potential. Conventional physicochemical synthesis methods often involve toxic chemicals, high energy consumption, and complex procedures that limit their clinical applicability and environmental safety. In contrast, biological synthesis utilizes microorganisms such as bacteria, fungi, and algae, as well as plant-derived phytochemicals, to reduce metal ions into stable nanoparticles under mild and eco-friendly conditions. Microbial-mediated synthesis relies on enzymatic reduction and metabolic pathways that enable controlled nanoparticle nucleation and growth, while phyto-extract mediated synthesis exploits bioactive compounds like flavonoids, phenolics, and terpenoids as natural reducing and capping agents. These green-synthesized nanoparticles exhibit enhanced surface functionalization, improved colloidal stability, and reduced cytotoxicity, making them highly suitable for biomedical applications. Recent studies demonstrate their effectiveness in antimicrobial therapy, anticancer treatment, wound healing, drug delivery, biosensing, and diagnostic imaging. Additionally, the inherent bioactivity of plant and microbial biomolecules further enhances therapeutic efficacy and targeted cellular interactions. Despite challenges related to standardization, large-scale production, and comprehensive toxicity evaluation, green synthesis remains a promising and sustainable strategy for next-generation nanomedicine. This review comprehensively discusses synthesis mechanisms, influencing parameters, characterization approaches, and diverse biomedical applications of microbially and phyto-derived metal nanoparticles.

Keywords: Green synthesis, Metal nanoparticles, Microbial-mediated synthesis, Phyto-extracts, Biomedical applications

Introduction

Green synthesis of metal nanoparticles (MNPs) using microbial and plant extracts has emerged as a sustainable and biocompatible alternative to conventional physicochemical synthesis methods. Traditional nanoparticle fabrication techniques often rely on toxic reducing agents, high energy consumption, and harsh reaction conditions that can compromise biological compatibility and environmental safety. In contrast, green synthesis employs natural biomolecules present in microorganisms (bacteria, fungi, algae) and plant tissues (leaves, roots, bark, fruits) as reducing, stabilizing, and capping agents to convert metal ions into nanoscale particles. These biomolecules—including polyphenols, flavonoids, terpenoids, proteins, enzymes, and polysaccharides—facilitate rapid reduction of metal salts such as Ag^{+}, Au^{3+}, Zn^{2+}, and Cu^{2+} into stable nanoparticles with controlled morphology and size distribution. The biological route not only minimizes hazardous by-products but also imparts inherent surface functionalization, enhancing colloidal stability and enabling direct integration into biomedical systems. Microbial-mediated synthesis often involves intracellular or extracellular enzymatic reduction mechanisms, whereas plant extract-mediated synthesis primarily relies on phytochemical redox reactions, both of which provide eco-friendly, cost-effective, and scalable approaches. Consequently, green-synthesized MNPs (G-MNPs) represent a convergence of nanotechnology and biotechnology aimed at producing functional nanomaterials with improved safety profiles and reduced ecological impact.

The biomedical relevance of green-synthesized metal nanoparticles is particularly significant due to their unique physicochemical properties, including high surface-area-to-volume ratio, tunable optical characteristics, and enhanced reactivity at the nanoscale. These attributes enable diverse therapeutic and diagnostic applications such as antimicrobial coatings, anticancer drug delivery, wound healing agents, biosensors, and imaging contrast materials. The bioactive phytochemicals or microbial proteins adsorbed on nanoparticle surfaces often confer additional pharmacological functionality, improving cellular uptake, reducing cytotoxicity, and promoting targeted interactions with biological macromolecules. For instance, silver and gold nanoparticles synthesized via plant extracts exhibit strong antimicrobial and anticancer activities through mechanisms involving reactive oxygen species generation, membrane disruption, and apoptosis induction. Similarly, microbially synthesized nanoparticles demonstrate enhanced enzyme-mimetic and immunomodulatory properties that are valuable in tissue engineering and regenerative medicine. Importantly, the green synthesis approach aligns with principles of green chemistry and sustainable nanomedicine by integrating renewable biological resources, mild reaction conditions, and minimal environmental footprint. As a result, the development of G-MNPs using microbial and plant extracts has gained substantial attention in translational biomedical research, offering promising prospects for next-generation therapeutic platforms that combine efficacy, safety, and ecological responsibility.

Fundamentals of Nanoparticles

Nanoparticles are defined as particulate materials with at least one dimension in the range of 1–100 nm, exhibiting unique physicochemical properties that differ significantly from their bulk counterparts due to quantum confinement effects, high surface-area-to-volume ratio, and

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enhanced surface reactivity. Metal nanoparticles (MNPs), including silver, gold, copper, and metal oxides, are particularly important in biomedical research owing to their tunable optical, electronic, catalytic, and magnetic properties. At the nanoscale, electrons are confined within discrete energy levels, leading to size-dependent optical phenomena such as surface plasmon resonance (SPR), which is widely exploited in biosensing and imaging. The high surface energy and large proportion of atoms at the nanoparticle interface facilitate strong interactions with biological molecules, enabling improved cellular uptake, targeted drug delivery, and antimicrobial efficacy. Key parameters governing nanoparticle performance include size distribution, morphology, crystallinity, surface charge, and functionalization, all of which influence colloidal stability and biological compatibility. Conventional synthesis methods—such as chemical reduction, sol–gel processing, and physical vapor deposition—allow precise control over these characteristics but often involve toxic reagents, high temperature, and substantial energy input. Such limitations raise concerns regarding environmental sustainability and biomedical safety. Consequently, the development of eco-friendly synthesis strategies that maintain control over nanoparticle physicochemical attributes while minimizing toxicity has become a major focus in nanobiotechnology. Understanding the fundamental principles governing nanoparticle formation, nucleation, growth kinetics, and stabilization is therefore essential for tailoring nanomaterials with optimized performance for therapeutic, diagnostic, and regenerative medicine applications.

Concept of Green Synthesis

Green synthesis refers to the eco-friendly fabrication of nanoparticles using biological entities or natural biomolecules as reducing, stabilizing, and capping agents, in alignment with the principles of green chemistry and sustainable nanotechnology. Unlike conventional physicochemical synthesis, which relies on hazardous solvents and strong reducing agents such as sodium borohydride or hydrazine, green synthesis utilizes renewable biological resources including plant extracts, microorganisms, enzymes, and polysaccharides to convert metal ions into stable nanoparticles under mild reaction conditions. The fundamental mechanism involves the bioreduction of metal precursors (e.g., Ag^{+}, Au^{3+}, Zn^{2+}) through redox-active phytochemicals or microbial metabolites, followed by nucleation and controlled growth of nanoparticles stabilized by natural capping biomolecules. These biomolecules not only prevent aggregation but also impart intrinsic surface functionalization, enhancing biocompatibility and therapeutic interaction with biological systems. The green synthesis paradigm emphasizes minimal energy consumption, aqueous solvent systems, and elimination of toxic by-products, thereby reducing environmental footprint and improving clinical safety profiles. Moreover, the approach enables facile scalability and cost-effectiveness, making it suitable for large-scale production of nanomaterials for biomedical use. The concept integrates interdisciplinary knowledge from

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chemistry, microbiology, and materials science to produce multifunctional nanostructures with enhanced pharmacological properties such as antimicrobial, antioxidant, and anticancer activities. Consequently, green synthesis is considered a transformative strategy for developing sustainable nanomedicine platforms that combine environmental responsibility with high therapeutic efficacy.

Literature Review

The green synthesis of metal nanoparticles (MNPs) using plant extracts has been extensively investigated as a sustainable alternative to conventional chemical and physical methods. Early foundational reviews emphasized the role of phytochemicals as natural reducing and stabilizing agents that facilitate nanoparticle formation under mild conditions. Singh et al. (2016) systematically explained that plant-mediated synthesis involves biomolecules such as flavonoids, terpenoids, alkaloids, and phenolic acids, which reduce metal ions and cap the resulting nanoparticles, ensuring stability and biocompatibility. Ahmed et al. (2016) further highlighted that plant extracts enable the synthesis of silver nanoparticles (AgNPs) with controlled morphology, improved antimicrobial activity, and reduced toxicity compared to chemically synthesized counterparts. Similarly, Iravani (2017) discussed how plant-based green nanotechnology aligns with green chemistry principles by minimizing hazardous reagents and energy-intensive processes. These works collectively established that phyto-mediated nanoparticle synthesis is eco-friendly, scalable, and capable of producing nanoparticles with diverse shapes and sizes, which are critical parameters influencing biomedical functionality. Moreover, the integration of naturally occurring bioactive compounds on nanoparticle surfaces enhances their pharmacological interactions, enabling applications in antimicrobial therapy, wound healing, and drug delivery. Thus, plant extract-based synthesis has emerged as a cornerstone in the development of sustainable nanomedicine.

Several empirical investigations have expanded upon these theoretical foundations by demonstrating specific biomedical applications of plant-mediated nanoparticles. Masurkar and Chaudhari (2017) reported the biosynthesis of silver nanoparticles using plant extracts and documented their significant antimicrobial and antifungal activities, attributing these effects to reactive oxygen species (ROS) generation and membrane damage in pathogens. Vijayakumar et al. (2018) synthesized gold nanoparticles using medicinal plant extracts and observed potent anticancer activity, suggesting that phytochemical-capped nanoparticles can induce apoptosis and inhibit tumor cell proliferation. Gondwal and Pant (2018) evaluated the antibacterial potential of plant-synthesized metal nanoparticles and confirmed their efficacy against multidrug-resistant bacterial strains. These studies collectively demonstrate that plant-derived nanoparticles not only act as passive nanocarriers but also exhibit intrinsic therapeutic properties due to the synergistic effect between metal cores and plant biomolecules. The biomedical

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relevance of such nanoparticles is further enhanced by their high surface reactivity, enabling targeted cellular interactions and improved bioavailability. Consequently, plant-based green synthesis is increasingly recognized as a multifunctional strategy that integrates nanoparticle fabrication with therapeutic efficacy, thereby advancing the field of biogenic nanomaterials.

Beyond plant-mediated approaches, microbial synthesis of metal nanoparticles has gained attention due to its mechanistic precision and enzymatic control over nanoparticle formation. Patra et al. (2017) emphasized that bacteria, fungi, and algae can reduce metal ions through intracellular and extracellular enzymatic pathways, producing nanoparticles with well-defined size distribution and crystallinity. Salem et al. (2022) elaborated on the role of microbial metabolites, proteins, and reductase enzymes in converting metal salts into stable nanoparticles suitable for biomedical applications. Microbial synthesis offers advantages such as scalability, reproducibility, and the ability to tailor nanoparticle properties through metabolic engineering. Furthermore, the microbial cell wall provides a natural template for nanoparticle nucleation and stabilization, enhancing structural uniformity and functional activity. This biosynthetic route has been successfully applied to produce silver, gold, zinc oxide, and copper nanoparticles with promising antimicrobial, antioxidant, and anticancer properties. The enzymatic reduction mechanisms involved in microbial synthesis also reduce the need for external chemical stabilizers, thereby minimizing toxicity and improving clinical applicability. These findings collectively highlight microbial-mediated green synthesis as a complementary and versatile platform for generating biocompatible nanomaterials with controlled physicochemical attributes.

Comprehensive reviews have also examined the interaction between biomolecules and nanoparticles, elucidating the mechanistic basis of their biological activity. Roy et al. (2019) discussed how proteins, polysaccharides, and phenolic compounds adsorbed onto nanoparticle surfaces modulate their physicochemical stability and biological interactions. Such biomolecule-nanoparticle conjugation enhances dispersion stability, prevents aggregation, and promotes targeted binding to microbial or cancer cell membranes. Kharissova et al. (2016) provided a broader analytical perspective, describing greener synthesis routes and emphasizing that the choice of biological source directly influences nanoparticle size, morphology, and functionality. These insights demonstrate that the green synthesis process is not merely a reduction reaction but a complex bio-nano interface phenomenon where biomolecular capping determines biological performance. Khan et al. (2019) further reviewed the physicochemical properties and toxicological considerations of nanoparticles, noting that biogenic nanoparticles generally exhibit lower cytotoxicity and improved safety profiles due to natural surface functionalization. Collectively, these studies underline the importance of understanding biomolecule-mediated stabilization and its implications for biomedical efficacy and biosafety.

In addition to mechanistic and application-oriented perspectives, several authors have provided integrative overviews of plant and microbial green nanotechnologies. Makarov et al. (2018) described plant-based "green" nanotechnologies as interdisciplinary platforms that combine nanoscience with plant biochemistry to produce eco-friendly nanomaterials. Their analysis emphasized that plant extracts serve as multifunctional agents capable of simultaneous reduction, capping, and functionalization of nanoparticles, which simplifies synthesis protocols and enhances biomedical compatibility. Al-Abodi and Hassan (2023) expanded this concept by reviewing different plant extracts used in nanoparticle synthesis and comparing their efficiency in producing stable and biologically active nanoparticles. They concluded that variations in phytochemical composition across plant species lead to differences in nanoparticle morphology, stability, and therapeutic performance. Such comparative analyses highlight the need for systematic optimization of plant selection and extraction parameters to achieve reproducible nanoparticle characteristics. Furthermore, the integration of ethnomedicinal plants in nanoparticle synthesis opens new avenues for developing nanotherapeutics that combine traditional medicinal properties with nanoscale advantages, thereby bridging conventional herbal medicine and modern nanotechnology.

The reviewed literature demonstrates a coherent evolution of green synthesis strategies from fundamental mechanistic studies to advanced biomedical applications. The convergence of plant-based and microbial biosynthesis approaches has enabled the production of diverse metal nanoparticles with enhanced antimicrobial, anticancer, antioxidant, and drug delivery capabilities. Foundational reviews established the theoretical principles of biomolecule-mediated reduction and stabilization, while experimental studies validated the therapeutic efficacy of biogenic nanoparticles in various biomedical contexts. Mechanistic investigations further clarified how surface-bound biomolecules regulate nanoparticle stability, reactivity, and cellular interactions, ultimately influencing biological outcomes. Despite these advances, challenges remain regarding large-scale production, reproducibility, and comprehensive toxicity evaluation, which are critical for clinical translation. Nevertheless, the accumulated evidence strongly supports green synthesis as a sustainable and versatile platform for fabricating functional nanomaterials. The integration of plant and microbial systems not only reduces environmental impact but also enhances the therapeutic potential of nanoparticles through natural biofunctionalization. Consequently, green-synthesized metal nanoparticles represent a promising frontier in nanomedicine, offering eco-friendly, biocompatible, and multifunctional solutions for next-generation biomedical applications.

Summary of Key Literature on Green Synthesis of Metal Nanoparticles for Biomedical Applications

S.	Author(s)	Source Type	Biological	Key Focus	Major	Biomedical

No.	& Year		System Used		Findings / Contributions	Relevance
1	Al-Abodi & Hassan (2023)	Review	Various plant extracts	Comparative review of plant-mediated nanoparticle synthesis	Demonstrated influence of phytochemical composition on nanoparticle size, stability, and activity	Antimicrobial, anticancer, and drug delivery potential
2	Masurkar & Chaudhari (2017)	Research/Review	Plant extracts (AgNPs)	Biosynthesis and biomedical uses of silver nanoparticles	Reported strong antimicrobial and antifungal activity due to ROS generation and membrane disruption	Antimicrobial coatings and wound healing
3	Vijayakumar et al. (2018)	Experimental Study	Medicinal plant extracts (AuNPs)	Anticancer activity of gold nanoparticles	Showed apoptosis induction and tumor growth inhibition	Cancer therapeutics and targeted therapy
4	Gondwal & Pant (2018)	Experimental Study	Plant extracts	Green synthesis and antibacterial evaluation	Confirmed efficacy against multidrug-resistant bacterial strains	Antibacterial agents and infection control
5	Singh et al. (2016)	Review	Plants and microorganis	Biological synthesis	Explained roles of	Broad biomedical

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			ms	mechanisms	phytochemicals and enzymes in reduction and stabilization of nanoparticles	nanotechnology applications
6	Iravani (2017)	Review	Plant systems	Green chemistry-based nanoparticle synthesis	Highlighted eco-friendly, cost-effective synthesis aligned with sustainable nanotechnology	Safe nanomedicine development
7	Ahmed et al. (2016)	Review	Plant extracts (AgNPs)	Green synthesis of silver nanoparticles	Discussed controlled morphology, enhanced stability, and reduced toxicity	Antimicrobial therapy and biosensing
8	Kharissova et al. (2016)	Review	Plants, microbes, biomolecules	Greener synthesis routes	Emphasized role of biological matrices in controlling nanoparticle morphology and functionality	Environmentally safe biomedical nanomaterials
9	Roy et al. (2019)	Review	Biomolecule-mediated synthesis	Biomolecule–nanoparticle interactions	Demonstrated how proteins and phenolics enhance stability and	Drug delivery and antimicrobial systems

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					biological targeting	
10	Salem et al. (2022)	Review	Bacteria and fungi	Microbial biosynthesis of nanoparticles	Described enzymatic reduction mechanisms and biomedical utility	Antimicrobial, antioxidant, and regenerative medicine
11	Khan et al. (2019)	Review	General nanoparticle systems	Properties, applications, and toxicity	Addressed physicochemical properties and safety concerns of nanoparticles	Risk assessment and safe clinical translation
12	Patra et al. (2017)	Review	Microorganisms (bacteria, fungi, algae)	Microbial nanoparticle synthesis	Highlighted enzymatic control, scalability, and uniform nanoparticle formation	Nanomedicine, biosensors, and therapeutics
13	Makarov et al. (2018)	Review	Plant-based green nanotechnologies	Plant-mediated nanoparticle synthesis	Showed plants act as reducing, capping, and functionalizing agents simultaneously	Eco-friendly nanotherapeutics and diagnostics

Microbial-Mediated Green Synthesis of Metal Nanoparticles

Microbial-mediated green synthesis of metal nanoparticles exploits the metabolic machinery of bacteria, fungi, algae, and actinomycetes to reduce metal ions into nanoscale particles through enzymatic and non-enzymatic pathways. In this biosynthetic route, microorganisms secrete reductase enzymes, proteins, peptides, and secondary metabolites that facilitate the conversion of metal salts into stable nanoparticles either intracellularly or extracellularly. Intracellular synthesis involves uptake of metal ions followed by enzymatic reduction within the cytoplasm, whereas extracellular synthesis occurs through secreted biomolecules that act as reducing and capping agents in the surrounding medium. Microbial cell walls rich in functional groups such as carboxyl, hydroxyl, and amine moieties provide nucleation sites for nanoparticle formation, enabling precise control over particle size, morphology, and crystallinity. Fungal systems are particularly advantageous due to their high biomass yield and abundant enzyme secretion, leading to efficient large-scale nanoparticle production. Microbial synthesis is reproducible, scalable, and capable of generating nanoparticles with uniform distribution and enhanced stability, making it suitable for biomedical applications including antimicrobial agents, biosensors, and targeted drug delivery systems. Additionally, metabolic engineering and genetic modification strategies can be employed to tailor nanoparticle characteristics by manipulating enzyme expression levels and metabolic pathways. This biologically controlled synthesis route minimizes the use of hazardous chemicals and produces nanoparticles with natural surface coatings that improve biocompatibility and reduce cytotoxicity. Hence, microbial-mediated synthesis represents a robust and versatile platform for producing functional nanomaterials for advanced biomedical applications.

Phyto-Extract Mediated Green Synthesis

Phyto-extract mediated green synthesis involves the utilization of plant-derived biomolecules to reduce metal ions and stabilize the resulting nanoparticles, offering a rapid, cost-effective, and environmentally benign synthesis pathway. Plant extracts obtained from leaves, roots, stems, bark, fruits, or seeds contain diverse phytochemicals such as flavonoids, phenolic acids, terpenoids, alkaloids, saponins, and reducing sugars, which act synergistically as reducing and capping agents during nanoparticle formation. Upon mixing plant extract with metal salt solutions, these biomolecules donate electrons to metal ions, leading to nucleation and subsequent growth of nanoparticles while simultaneously adsorbing onto their surfaces to prevent aggregation and provide colloidal stability. The composition and concentration of phytochemicals, along with reaction parameters such as pH, temperature, and extract-to-metal ion ratio, critically influence nanoparticle size, shape, and dispersity. Phyto-mediated synthesis is particularly advantageous due to its simplicity, rapid reaction kinetics, and elimination of the need for microbial culture maintenance or sterile conditions. Furthermore, plant-capped nanoparticles often inherit the bioactive properties of the parent phytochemicals, resulting in enhanced antimicrobial, antioxidant, anti-inflammatory, and anticancer activities. This intrinsic biofunctionalization improves cellular compatibility and facilitates targeted therapeutic interactions. Consequently, phyto-extract mediated synthesis has become one of the most widely

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adopted green approaches for fabricating metal nanoparticles intended for biomedical applications, bridging traditional herbal biochemistry with modern nanomedicine.

Conclusion

Green synthesis of metal nanoparticles using microbial and phyto-extracts represents a transformative and sustainable paradigm in nanobiotechnology with significant implications for biomedical applications. By harnessing the intrinsic reducing and stabilizing capabilities of microbial metabolites and plant-derived phytochemicals, this approach enables the eco-friendly fabrication of nanoparticles with controlled size, morphology, and enhanced surface functionality. Compared with conventional physicochemical methods, biologically synthesized nanoparticles exhibit superior biocompatibility, reduced toxicity, and improved colloidal stability, making them more suitable for clinical and therapeutic applications. Microbial-mediated synthesis offers advantages such as enzymatic precision, scalability, and reproducibility, while phyto-extract mediated synthesis provides rapid, cost-effective, and versatile routes driven by diverse bioactive compounds. These complementary biosynthetic strategies have demonstrated remarkable efficacy in antimicrobial therapy, anticancer treatment, wound healing, targeted drug delivery, and biosensing technologies, highlighting their multifunctional biomedical potential.

Despite these promising advancements, several challenges remain, including variability in biological extract composition, lack of standardized synthesis protocols, and insufficient long-term toxicity and in vivo validation studies. Addressing these limitations requires systematic optimization of reaction parameters, mechanistic understanding of biomolecule–nanoparticle interactions, and development of scalable production frameworks compliant with regulatory standards. Future research should focus on integrating omics-driven microbial engineering, advanced characterization tools, and translational clinical evaluations to fully harness the therapeutic potential of green-synthesized nanoparticles. Overall, the convergence of green chemistry, microbiology, and nanomedicine positions microbial and phyto-mediated nanoparticle synthesis as a key platform for developing safe, sustainable, and multifunctional nanotherapeutics for next-generation biomedical innovations.

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