Biological Trace Element Research (2024) 202:3383–3399
https://doi.org/10.1007/s12011-023-03920-9

RESEARCH

Recent Trends in Biologically Synthesized Metal Nanoparticles and their Biomedical Applications: a Review

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Received: 1 August 2023 / Accepted: 14 October 2023 / Published online: 25 October 2023

Abstract

In recent years, biologically synthesized metal nanoparticles have emerged as a dynamic field of research with significant implications for biomedical applications. This review explores the latest trends in the synthesis of metal nanoparticles using biological methods, encompassing plant extracts and microorganisms such as bacteria, yeasts, and fungi. These innovative approaches offer a sustainable, cost-effective, and environmentally friendly alternative to conventional chemical synthesis methods. Moreover, this review delves into the multifaceted biomedical applications of biologically synthesized metal nanoparticles. These applications include drug delivery systems, diagnostics, therapeutics, and imaging technologies, showcasing the versatility and promise of these nanomaterials in addressing contemporary biomedical challenges. In addition, the review addresses the critical issue of cytotoxicity, offering insights into the safety and viability of these biologically derived NPs for medical use. The exploration of recent trends and advancements in this field underscores the transformative potential of biologically synthesized metal nanoparticles in revolutionizing biomedical research and healthcare.

Keywords Biological synthesis · Metallic nanoparticles · Biomedical application · Cytotoxicity

Introduction

Nanotechnology has ushered in a new era of innovation across various scientific domains, and one of its most intriguing frontiers is the synthesis of metal nanoparticles (MNPs). These nanoscale materials exhibit distinctive physical, chemical, and biological properties due to their reduced dimensions, making them highly attractive for a wide range of applications [1]. Transitioning these innovations from laboratory settings to practical use is the essential step towards their real-world impact. The commercialization of these groundbreaking technologies is vital for a broad spectrum of human needs and global advancement [2]. However, meticulous attention must be paid to materials' potential, health assessments, and environmental consequences. It is undeniable that nanoparticles (NPs) pose health concerns that demand swift attention. Moreover, their production and utilization remain largely unregulated, particularly in the context of a rapidly evolving industrial landscape [3]. As new chemical processes are designed with minimal risks in mind, hazardous compounds are minimized or eliminated by adhering to a set of fundamental principles. This underscores the critical role of the burgeoning green chemistry industry [4].

Significant time and effort have been invested in developing viable synthetic methodologies for producing nanoparticles, primarily due to their distinctive physicochemical properties and wide-ranging applications. Nevertheless, the environmental hazards stemming from heavy metal residues have limited the utility of numerous physicochemical techniques in nanoparticle production. Consequently, the adoption of biological approaches for nanoparticle synthesis has emerged as a promising trend in the industry. This shift is driven by several key advantages, including non-toxicity,

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reproducibility, ease of scalability, and the ability to achieve well-defined nanoparticle shapes [3–5].

In recent years, there has been a growing emphasis on harnessing the capabilities of biological systems for the synthesis of MNPs, resulting in the emergence of biologically synthesized metal nanoparticles (bio-MNPs). The convergence of nanotechnology and biotechnology has paved the way for innovative and eco-friendly methods of producing MNPs. Employing "green synthesis" techniques is crucial to minimize the generation of undesirable or hazardous by-products and to promote the development of dependable, sustainable, and environmentally friendly synthesis methods [6]. Biogenic synthesis, involving the use of microorganisms, plants, and other natural resources, offers a sustainable approach that not only mitigates the environmental impact associated with traditional chemical methods but also provides precise control over the properties of the synthesized nanoparticles. As a result, bio-MNPs have garnered considerable attention across scientific disciplines, with biomedical applications taking center stage [7].

The biomedical field is witnessing a transformative paradigm shift, with bio-MNPs gaining prominence as versatile tools for various diagnostic, therapeutic, and imaging applications. Their unique physicochemical properties, surface functionality, and biocompatibility make them ideal candidates for targeted drug delivery, biosensing, cancer therapy, and bioimaging. The ability to tune the size, shape, and surface characteristics of bio-MNPs adds a layer of customization that is crucial for optimizing their interactions with biological systems [2, 7].

In this context, we provide an update on recent breakthroughs in the synthesis of biological nanoparticles and offer insights into their potential applications and future prospects.

Biological Synthesis of Metallic Nanoparticles

Biological synthesis of metallic nanoparticles refers to the process of creating nanoscale metallic particles using natural sources, such as microorganisms (bacteria, fungi, yeast) and plants. This approach, often referred to as "green synthesis," offers an environmentally friendly and sustainable alternative to traditional physicochemical methods that may involve toxic chemicals or generate harmful by-products [8]. During the biological synthesis of metallic nanoparticles, biological agents or extracts from these agents are used to reduce and stabilize metal ions from a precursor solution, leading to the formation of nanoparticles with controlled sizes and shapes [6].

This method has gained significant attention due to its numerous advantages, including [4, 7–9]:

Non-toxicity Biological synthesis utilizes mild reaction conditions and natural reducing agents, minimizing the use of hazardous chemicals and reducing toxicity in the final product.

Eco-friendliness The process often involves readily available biological materials, which reduces the environmental impact associated with traditional chemical processes.

Control over Size and Shape By adjusting reaction parameters and utilizing specific biological agents, it is possible to control the size and morphology of the resulting nanoparticles.

Reproducibility Biological synthesis methods can be standardized, leading to consistent and reproducible nanoparticle production.

Biocompatibility The resulting nanoparticles often exhibit better biocompatibility, making them suitable for various biomedical applications.

Scalability Some biological synthesis methods are easily scalable, allowing for large-scale production of nanoparticles.

Versatility Various microorganisms and plant extracts can be employed to synthesize nanoparticles of different metals, offering a versatile approach.

A pictorial demonstration of the biological synthesis of metallic nanoparticles is presented in Fig. 1.

Mechanisms of Biological Synthesis of Metallic Nanoparticles

The biological synthesis of metallic nanoparticles (MNPs) involves intricate biochemical and biophysical mechanisms that vary depending on the organism or biomolecule being used. Understanding these mechanisms is essential for tailoring the properties of MNPs for specific applications. Researchers continue to explore and optimize these processes to enhance the efficiency, reproducibility, and scalability of biological synthesis methods for MNPs [10]. Some common mechanisms involved in the biological synthesis of MNPs are:

1. Reduction of metal ions [10, 11]:

- *Enzymatic reduction*: In many biological synthesis methods, enzymes play a crucial role as reducing

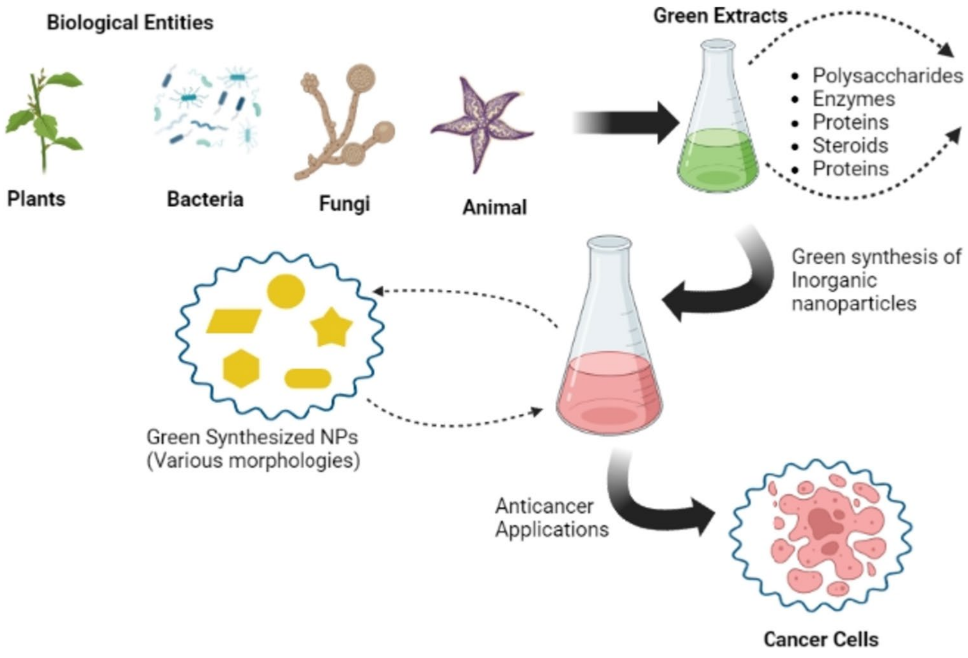


Fig. 1 Biological synthesis of metallic nanoparticles

agents. Enzymes like nitrate reductase, laccase, and chloroperoxidase can catalyze the reduction of metal ions (e.g., Ag^+, Au^{3+}) by providing electrons from their cofactors, co-substrates, or active sites. This reduction process leads to the formation of metal nanoparticles.

- *Microbial reduction*: Some microorganisms, such as bacteria and fungi, are capable of reducing metal ions extracellularly. These microorganisms produce specific biomolecules or metabolites (e.g., NADH) that serve as electron donors, facilitating the reduction of metal ions to MNPs.

2. Nucleation and growth [12]:

- *Nucleation:* Once metal ions are reduced, they tend to nucleate, forming small clusters of atoms. Nucleation is a critical step that determines the size and number of nanoparticles formed. Various factors, including temperature, pH, and concentration of metal ions, influence nucleation.
- *Growth:* After nucleation, the nanoparticles continue to grow by the addition of metal ions onto their surfaces. The rate of growth and the shape of the nanoparticles can be controlled by adjusting the reaction conditions.

3. Stabilization and capping [13]:

- *Biomolecule interaction:* Biomolecules present in the biological system, such as proteins, peptides, or polysaccharides, can interact with the newly formed MNPs. These biomolecules can serve as stabilizing agents, preventing the aggregation and precipitation of MNPs.
- *Surface modification:* The biomolecules that attach to the MNP surface can also affect the surface properties, such as charge and reactivity. This surface modification can be utilized to tailor the MNPs for specific applications.

4. Size and shape control [12, 14]:

Biological templates: Some biological systems provide templates or scaffolds for the formation of MNPs with specific shapes. For instance, viruses and plant proteins have been used to synthesize MNPs with well-defined shapes.

5. Intracellular vs. extracellular synthesis [11, 14]:

- *Intracellular synthesis:* In some cases, MNPs are synthesized within the cells of microorganisms. These intracellularly synthesized MNPs can be stored or excreted by the organisms.
- *Extracellular synthesis:* In other instances, MNPs are formed outside the cells in the extracellular environment. This extracellular synthesis method is commonly used in bio-MNP production.

6. pH and temperature dependence [13]:

- *pH influence:* pH plays a critical role in biological synthesis as it affects the redox potential and charge of metal ions, impacting their reduction and subsequent nanoparticle formation.
- *Temperature effects:* Temperature can influence reaction rates and the stability of formed MNPs. Optimizing temperature conditions is essential for controlling MNP synthesis.

Plant-Based Synthesis of Metallic Nanoparticles

Plant nanotechnology has recently unveiled innovative pathways for the eco-friendly production of nanoparticles, offering a straightforward, rapid, and stable approach. Employing water as a reducing agent in nanoparticle synthesis brings numerous advantages, including biocompatibility, scalability, and applicability in medicine. This signifies that nanoparticles derived from plant sources hold the potential to meet the growing demand in biomedicine and environmental applications, primarily due to their accessibility and non-hazardous nature [14, 15]. Recent investigations have revealed the feasibility of producing gold and silver nanoparticles utilizing *Panax ginseng* leaf and root extracts, showcasing the potential of medicinal plants as viable sources of raw materials [16]. Furthermore, various plant components such as leaves, fruits, and stems, as well as their extracts, have been harnessed for the synthesis of metal nanoparticles.

The role of secondary metabolites, including flavonoids and alkaloids, has garnered attention for their pivotal roles in metal salt reduction, as well as their capability to serve as capping and stabilizing agents for nanoparticles produced from proteins and amino acids [17, 18]. For instance, *Corallina officinalis* extract, enriched with polyphenols and proteins bearing carbonyl groups, has exhibited potential in facilitating the generation and stabilization of gold nanoparticles [19].

Comparing biologically synthesized nanoparticles with their physicochemically synthesized counterparts, literature suggests that biogenic nanoparticles tend to exhibit heightened activity. Achieving stable and monodisperse metallic nanoparticles necessitates accurate control over pH, incubation time, mixing ratios, and temperature. Diverse plants, including curry, mango, neem, turmeric, and guava, have been explored for the creation of gold nanoparticles. These plant extracts, rich in polyphenols, expedite the breakdown of organic matter, further influencing nanoparticle formation [20]. Moreover, plants demonstrate an extraordinary capacity to reduce metal ions, not only on their surfaces but also in distant organs and tissues. This phenomenon has led to the prevalence of nanoparticles (NPs) as the primary form of metal deposition, as evidenced by studies on metal bioaccumulation [21]. For instance, the leaf extract of *Diopyros kaki* was employed as a green and eco-friendly reducing agent in the extracellular synthesis of platinum nanoparticles from an aqueous $H_{(2)}PtCl_{(6)}0.6H_{(2)}O$ solution [22]. Similarly, *Murraya Koenigii* leaf extract served a dual role as both a reducing and stabilizing agent, facilitating the production of silver nanoparticles at ambient conditions and gold nanoparticles at 373 K. The resulting nanoparticles underwent thorough characterization through UV–vis, transmission electron microscopy (TEM), X-ray diffraction (XRD), and FTIR analysis. This innovative method yielded well-dispersed silver nanoparticles with a size of approximately 10 nm and gold nanoparticles with a size of approximately 20 nm [23]. Notably, the concentration of the plant extract has been demonstrated to exert a significant influence on the reduction rate, particle size, and propensity for agglomeration.

As evidenced by these diverse studies, the realm of plant nanotechnology holds immense promise in revolutionizing nanoparticle synthesis. Through the strategic utilization of plant constituents, researchers are uncovering innovative pathways for the sustainable production of nanoparticles with a wide array of potential applications [24].

Animal-Based Synthesis of Metallic Nanoparticles

Animal-based synthesis of metallic nanoparticles involves using various animal-derived substances or components to facilitate the production of metallic nanoparticles. Researchers working on these methods aim to harness the natural properties of animal-derived materials to create metallic nanoparticles for various purposes, including biomedical, catalytic, and environmental applications [25]. While the majority of nanoparticle synthesis methods are plant-based or microbial, there are some examples where animal-based materials are utilized for this purpose. A range of natural sources and marine organisms have been explored for the synthesis of nanomaterials, including fibroin-titanium dioxide nanocomposites, nano-hydroxyapatite (a form of calcium NPs), and other nanoparticles. These eco-friendly synthesis methods utilize silk proteins, derived from various species such as spiders, silkworms, and tree ants [26]. Sericin, another silk protein, has also proven effective in nanoparticle production. Marine organisms possessing enzymatic capabilities, such as sponges and starfish, have been found to produce biological silica nanoparticles. Earthworm extracts, notably from species like *Eisenia andrei*, have been employed for the creation of nano-red gold, particularly when trichlorogold hydrochloride is present. In the green synthesis of silver nanoparticles, marine worms like Polychaeta have shown promise [27]. Additionally, chitins obtained from crustacean shells, including chitosan, offer a valuable resource for the development of nanocomposites involving titanium dioxide and chitosan.

Microbiome-Based Synthesis of Metallic Nanoparticles

Microbiome-based synthesis of metallic nanoparticles involves the use of microorganisms, such as bacteria or fungi, and their associated microbiomes to facilitate the production of metallic nanoparticles. This approach leverages the unique metabolic capabilities of microorganisms and their interactions with the surrounding microbiota to synthesize nanoparticles with various properties [28]. Microbes can be used as safe and cheap tools for synthesis of metallic NPs like gold, silver, copper, zinc, titanium, palladium, and nickel. The synthesis of NPs can be carried out both extracellularly and intracellularly using microbes [29, 30].

Recently, bacterial strains belonging to *Acinetobacter calcoaceticus*, *Bacillus amyloliquefaciens*, *Bacillus megaterium*, *Bacillus licheniformis*, *Escherichia coli*, *Lactobacillus* sp., and *Pseudomonas stutzeri* have been used in for the biosynthesis of AgNPs [31, 32]. Zinc oxide NPs were synthesized using a bacterium *Aeromonas hydrophila* in simple and cost-effective method. The crystalline nature of the NPs was observed by atomic force microscopy (AFM), which showed that the NPs were spherical and oval with an average size of 57.72 nm [33].

Fungi are excellent sources of many bioactive compounds that can be utilized in various sectors. The fungal synthesis of metallic NPs is dependent on culture conditions. In a previous study, the culture conditions of *Trichothecium* sp. reduced Au ions resulting extracellular NPs synthesis but produced NPs intracellularly when cultured with agitations [34]. In another study, the biosynthesis of AgNPs using *Fusarium oxysporum* was reported where the effect of substrate concentration and incubation temperature was studied [35].

Yeast cells act as one of the most important agents for bioremediation of heavy metals. Yeasts are easily cultured in low-cost media and capable of removing various heavy metals. Saccharomyces cerevisiae was used for biosynthesis of AgNPs by biotransformation. Both the dried and fresh culture *S. cerevisiae* was used as the biocatalyst [36]. *Pichia jadinii* was used for intracellular synthesis of AuNPs ranging from 1 to 100 nm. In this study, the growth and cellular activities of *P. jadinii* were controlled easily to regulate AuNPs size and shape [37].

Several studies illustrate that viruses are considered to be a suitable group which serves as a biotemplate for material synthesis at the nanoscale to microscale [38]. Recently, material science researchers have been using the viral NPs (VNPs) as templates or scaffolds for the synthesis of novel hybrid nanomaterials [39]. The synthesis of nanomaterials using viruses is a clean, nontoxic, and environmentally friendly method which provides a broad range of sizes, shapes, compositions, and physicochemical properties [40]. In a study, a notorious plant pathogenic virus, squash leaf curl China virus (SLCCNV) was used as biotemplate to fabricate silver and gold nanomaterials. The SLCCNV was exposed to $HAuC_{l4}$ and $AgNO_3$ precursors in presence of sunlight and quick (~5 min) formation of SLCCNV-metallic-hybrid nanomaterials in an eco-friendly way was observed [41]. In another research, tobacco mosaic virus and bovine papilloma virus were used as additive materials with plant extracts *Avena sativa*, H*ordeumvulgare*, *Musa pradisiaca*, and *Nicotiana benthamiana*. These two viruses promoted the reduction and increase of the NPs number remarkably as compared to a control without virus [42]. These viral synthesized nanomaterials have a wide range of applications in biomedicine and serve as catalysts to biosensors [43].

Development of Biosynthesized Metallic Nanoparticles

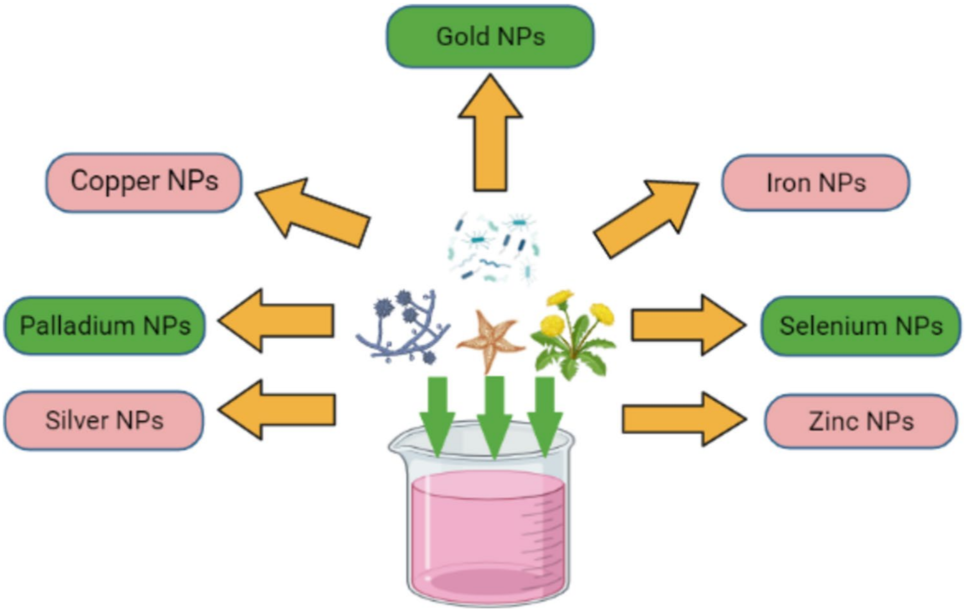
Biosynthesized metallic nanoparticles are nanoparticles that are produced through biological methods using various microorganisms, plants, and other biological entities as reducing and stabilizing agents. These nanoparticles are often referred to as “biogenic nanoparticles” or “green nanoparticles” due to their eco-friendly and sustainable production process [16, 20, 23]. The metallic nanoparticles developed using biological methods are presented in Fig. 2.

Gold Nanoparticles

Gold nanoparticles, comprising gold atoms, encompass a diverse range of sizes, spanning from a few nanometers to several hundreds of nanometers in diameter. These nanoparticles manifest distinctive characteristics owing to their diminutive dimensions and impressive surface area-to-volume ratio, setting them apart from bulk gold properties. The manifold attributes of gold nanoparticles render them invaluable across various domains, continually unveiling novel applications for exploration [44]. Extensive research has been devoted to exploring nanoparticle-based delivery systems for enhancing the loading of organic molecules. Particularly within the realm of nanotechnology, these particulate systems necessitate the incorporation of proteins, lipids, and carbohydrates. Notably, nanoparticles represent colloidal systems with size dimensions typically ranging from 10 to 1000 nm. [45, 46].

Gold, with its exceptional optical absorption properties, stands as an ideal candidate for detection purposes, while its photothermal attributes render it potent as an anticancer agent. Complex gold nanostructures can be meticulously

Fig. 2 Development of metallic nanoparticles by green synthesis method



engineered to optimize drug release efficacy. Strategies encompass the attachment of drug particles onto metal nanoparticle surfaces and the creation of hollow inner structures, augmenting entrapment efficiency. These structures are often easily modifiable to incorporate diverse optical properties. Furthermore, the nanoparticle surface can be coated with thermosensitive monomers to facilitate controlled drug delivery. These polymers enable the manipulation of drug-loaded particle suspension diffusion rates by contracting in warm conditions and expanding in colder environments. This innovative approach can be synergistically combined with gold's photothermal characteristics, offering a novel avenue for drug administration. For example, by focusing a laser on tumor sites to induce the melting of gold nanoparticles in proximity to the tumor location, effective drug delivery can be enhanced, while minimizing nonspecific toxicity.

Gum acacia (GA) has been employed as a polysaccharides-based technique to successfully establish gemcitabine hydrochloride (GEM)-loaded colloidal gold nanoparticles [47–49]. The improved water solubility and drug release rate of GEM when delivered on a gold carrier can be attributed to colloidal GEM-GA-AuNPs. Consequently, GEM's targeting effectiveness is significantly enhanced, owing to substantial increases in particle size, enhanced water solubility, and an improved drug release profile [47].

Silver Nanoparticles

Silver nanoparticles (AgNPs) stand as one of the most significant and captivating nanomaterials, finding versatile applications in various biomedical contexts. Within this domain, the environmentally friendly green synthesis of AgNPs using herbal extracts has garnered substantial attention, presenting an eco-conscious alternative. Notably, *Impatiens balsamina*, *Lantana camara*, and *Cucumis prophetarum* leaf extracts have been harnessed, demonstrating dual roles as both reducing and capping agents in the biosynthesis of AgNPs [50, 51]. Another noteworthy example revolves around the utilization of *Salvia officinalis*, a member of the salvia genus, renowned for its capacity to yield AgNPs imbued with remarkable antibacterial, antiplasmodial, antioxidant, anti-inflammatory, and antileishmanial properties [52, 53]. Intriguingly, geranium leaf extract has been employed in the extracellular synthesis of AgNPs. This process yields quasi-linear superstructures, spanning a size range from 16 to 40 nm, as evidenced by transmission electron microscopy (TEM). These AgNPs exhibit exceptional stability and crystallinity [54].

Further research has explored the potential of five plant leaf extracts (Pine, Persimmon, Ginkgo, Magnolia, and Platanus) in the extracellular synthesis of metallic silver nanoparticles. The plant leaf extracts served as effective reducing agents for Ag(+) to Ag(0) in aqueous $AgNO_3$ solutions, yielding stable silver nanoparticles. UV–visible spectroscopy was employed to quantitatively monitor the formation of these nanoparticles [55].

Additionally, *Azadirachta indica* leaf extract has been investigated as a potent reducing agent for the rapid biosynthesis of AgNPs. This research delved into the effects of AgNPs on growth, glutathione-S-transferase (GST) activity, total protein concentration, and potent antibacterial efficacy against *Staphylococcus aureus* [56]. The synthesis of AgNPs has also witnessed innovation through the combined use of *Mentha X Piperita* (Mint) and *Ocimum tenuiflorum* (Tulsi). This combination yielded AgNPs with outstanding antimicrobial activity, particularly against *Streptococcus mutans* [57].

Recent research has extended the green synthesis frontier by employing *Mangifera indica* leaf extracts to synthesize Mi-AgNPs. These Mi-AgNPs were found to be polycrystalline, spherical in shape, with an average size of 62 ± 13 nm [58].

Palladium Nanoparticles

Palladium nanoparticles (PdNPs) have garnered significant attention from researchers owing to their wide-ranging applications. Nonetheless, conventional methods for synthesizing PdNPs often entail the use of harmful solvents and reducing agents, leading to the generation of toxic pollutants and by-products [59]. To address these pressing environmental concerns, the green synthesis of palladium nanoparticles using plant extracts has emerged as a promising and sustainable alternative. This biosynthesis approach harnesses the inherent eco-friendliness of plant extracts, circumventing the drawbacks associated with traditional methods. Notably, these biologically synthesized nanoparticles exhibit advantageous characteristics, including enhanced selectivity and catalytic properties, rendering them highly appealing for diverse applications [60].

A compelling instance of this green synthesis involves the use of aqueous *Saccharomyces cerevisiae* extract for the biofabrication of palladium nanoparticles (PdNPs) and their subsequent photocatalytic applications [61]. Furthermore, the extract of *Diospyros kaki* leaves has been employed as a bio-stimulator to synthesize PdNPs, which demonstrated proficient antibacterial efficacy against both *Escherichia coli* and *Staphylococcus aureus*, yielding zones of inhibition measuring 18 and 10.5 mm, respectively [62]. Another noteworthy study showcased the feasibility of biosynthesizing Pd-NPs using *Shewanella loihica* PV-4, with a specific focus on their potential application as efficient catalysts for the remediation of chromium contamination [63].

Copper Nanoparticles

Copper nanoparticles have attracted significant attention among researchers due to their unique properties and cost-effectiveness, setting them apart from noble metal nanoparticles. In recent times, the green biosynthesis approach has gained notable prominence, particularly within the realm of biological applications. This preference arises from its simplicity, non-toxicity, and eco-friendly attributes, which stand in stark contrast to conventional physical and chemical synthesis methods. While some green materials exhibit dual functionality, serving as both stabilizing and reducing agents in the production of copper nanoparticles (Cu-NPs), others may necessitate an additional environmentally friendly boost [64, 65]. In a noteworthy study, copper nanoparticles (CuNPs) were successfully synthesized by treating an aqueous solution of copper sulfate pentahydrate with an extract derived from *Magnolia kobus* leaves, showcasing the potential of this green approach [66]. Additionally, stable copper nanoparticles were effectively synthesized through the utilization of *Ocimum sanctum* leaf extract, further exemplifying the eco-friendliness of green methods [67]. Furthermore, CuNPs were biosynthesized employing a *Pseudomonas stutzeri* bacterial strain isolated from wastewater originating from the electroplating industry, highlighting the adaptability of green synthesis techniques to various contexts [68].

In a recent investigation, the utilization of the *Stenotrophomonas maltophilia* strain SCS1.1 for the biosynthesis of copper oxide nanoparticles (CuONPs) proved significant. These CuONPs were subsequently evaluated for their inhibitory effects against various plant pathogenic microorganisms, showcasing the potential applications of green synthesized nanoparticles in agriculture and environmental contexts [69].

Iron Nanoparticles

Iron oxide nanoparticles (FeNPs) are widely employed due to their low toxicity and significant utility in everyday applications. In nature, two types of iron oxides, hematite and magnetite, hold distinct roles in scientific investigations [70]. Notably, iron oxide nanoparticles within the size range of 10–100 nm have played a pivotal role in various studies [71].

The utilization of *Ziziphora clinopodioides* leaf extract as a reducing agent has proven effective in the green synthesis of FeNPs from $FeCl_3$ solution. These FeNPs exhibited remarkable enhancements in immunological, hematological, and biochemical markers in anemic wild mice. Moreover, they demonstrated potent antioxidant activity against DPPH in vitro [72]. Additionally, FeNPs displayed significant dose-dependent cell viability effects in the HUVEC cell line, indicating their potential biomedical applications [56].

In another investigation, FeNPs were biosynthesized using *Phoenix dactylifera* extract as a reducing agent and iron sulfate heptahydrate ($FeSO_4 \cdot 7H_2O$) as a substrate. The antimicrobial activity of these synthesized FeNPs was assessed against various bacterial strains, including *Escherichia coli*, *Bacillus subtilis, Micrococcus leutus*, and *Klebsiella pneumoniae*. The results revealed differential antimicrobial potential, with the maximum zone of inhibition observed against Escherichia coli [73]. Furthermore, a separate study employed *Mentha spicata* for the synthesis of FeNPs and investigated their antimicrobial efficacy against *Phytophthora infestans* [74].

Zinc Oxide Nanoparticles

Zinc oxide nanoparticles (ZnO-NPs) stand as a prime example of metal oxide nanomaterials, valued for their unique physical and chemical properties [75]. These nanoparticles have captured the interest of researchers, with numerous studies utilizing various plant sources for their synthesis. For instance, the leaf extract of *Calotropis gigantea*, in the presence of $ZnNO_3$ salt, successfully yielded nanoparticles with a size range of 30–35 nm [76]. Functionalization of these green synthesized ZnONPs has revealed their potential in various domains. These nanoparticles exhibit promising anti-diabetic properties, high biocompatibility, and significant antibacterial activity [57]. Utilizing organic constituents present in *Wattakaka volubilis* leaf extract, ZnONPs were produced through reduction and capping of zinc ions. Beyond their anti-diabetic applications, these nanoparticles possess safety advantages, making them suitable for reliable pharmaceutical formulations and scalability. This development aligns with the evolution of drug design, emphasizing the need to identify and extract the active components responsible for their anti-diabetic effects [77, 78].

In a separate study, iron nanoparticles were synthesized using aqueous *Ageratum conyzoides* extracts, and their antimicrobial and photocatalytic properties were evaluated. Comprehensive characterization involved UV–Vis spectrophotometry, FT-IR spectrophotometry, X-ray diffractometry, and scanning electron microscopy. Gas chromatography-mass spectrometry (GC–MS) profiling of the extracts revealed the presence of secondary metabolites, with quantification of total phenolic and total flavonoids content. Antimicrobial activity was assessed against five microorganisms, demonstrating moderate activity compared to the antibiotic ciprofloxacin. Additionally, the nanoparticles exhibited photocatalytic efficiency in degrading methylene blue, achieving a degradation efficiency of 92%. These findings underscore the role of *Ageratum conyzoides* in the bioreduction of iron ions to FeNPs, with potential applications in microorganism control and photocatalytic processes [79].

Platinum Nanoparticles

Platinum nanoparticles (PtNPs) represent invaluable scientific tools, under intense exploration across various biotechnological, nanomedical, and pharmacological domains. Their remarkable attributes, including antimicrobial, antioxidant, and anticancer properties, have spurred extensive research [80, 81].

In one notable study, platinum nanoparticles were synthesized using *Ocimum sanctum* leaf extract as a reducing agent in the presence of aqueous chloroplatinic acid ($H_2PtCl_6H_2O$). Leveraging an *O. sanctum* leaf broth at a reaction temperature of 100 °C resulted in a more efficient conversion of platinum ions into nanoparticles [82]. In another investigation, *Atriplex halimus* leaf aqueous extract played a pivotal role in the green synthesis of platinum nanoparticles (At-PtNPs). These At-PtNPs exhibited remarkable stability over a span of 3 months. They served as highly effective catalysts for the degradation of MB dye and displayed significant antibacterial efficacy against Gram-negative bacteria. Additionally, At-PtNPs showcased potent antioxidant properties [83]. These findings underscore the versatile applications and promising potential of platinum nanoparticles in various scientific and biomedical endeavors.

Selenium Nanoparticles

Selenium has garnered increased attention in recent years due to its vital role in human health. This essential element is involved in critical metabolic pathways, including thyroid hormone regulation and immune system function. Selenium also plays a key role in safeguarding cells from damage caused by free radicals, as it facilitates the incorporation of antioxidant enzymes [84].

In a noteworthy study, selenium nanoparticles, known as phytofabricated selenium nanoparticles (PF-SeNPs), were synthesized using the aqueous fruit extract of *Emblica officinalis* through a simple, green, cost-effective, and environmentally friendly method. The fruit extract of *E. officinalis* was found to be rich in secondary metabolites such as phenolics, flavonoids, and tannins, rendering it highly suitable for nanoparticle biosynthesis. PF-SeNPs exhibited remarkable antioxidant, antimicrobial, and biocompatible properties. These nanoparticles displayed a wide range of antimicrobial activity, with fungi being the most susceptible, followed by Gram-positive and Gram-negative bacteria. Importantly, cytotoxicity studies revealed that PF-SeNPs were significantly less toxic and safer than sodium selenite [85].

In another study, selenium nanoparticles (SeNPs) were synthesized using extracts from three distinct plants: *Allium cepa* (onion), *Malpighia emarginata* (acerola), and *Gymnanthemum amygdalinum* (boldo). Antibacterial activity was assessed using microdilution in broth, followed by a time-kill curve analysis against bacterial strains, including *Staphylococcus aureus*, methicillin-resistant *S. aureus*, *Pseudomonas aeruginosa*, and *Escherichia coli* (Tables 1 and 2). The observed antimicrobial efficacy and low hemolytic concentration suggest the potential utility of these nanoparticles against Gram-positive bacteria, including multidrug-resistant strains, thereby opening up a wide range of applications [115].

Biomedical Application of Biosynthesized Metallic Nanoparticles

Metallic nanoparticles (MNPs) find extensive utility across various biomedical applications, encompassing medical diagnostics, molecular biology, bioimaging, drug

Table 1 Synthesis of metallic nanoparticles by different plant extracts

Plant extracts	Type of nanoparticles	Location	Size range (nm)	References
Azadirachta indica	Ag, Au, and Ag/Au	Extracellular	50–100	[86]
Pelargonium graveolens	Ag	-	16–40	[87]
Cymbopogon citratus	Au	-	200–500	[88]
Avena sativa (oat)	Au	Extracellular	5–85	[89]
Alfalfa sprouts	Ag	Intracellular	2–20	[90]
Aloe vera	Au	Extracellular	50–350	[91]
Cinnamomum camphora	Au and Ag	Extracellular	55–80	[92]
Asparagus racemosus	Pt/Pd NPs	-	1–6	[93]
Ginkgo biloba	Cu	-	60	[94]

Table 2 Synthesis of metallic nanoparticles by different microorganisms

Microorganism	Type of nanoparticles	Location	Size range (nm)	References
(A) Bacteria				
Pseudomonas stutzeri	Ag	Intracellular	~200	[95]
Morganella sp.	Ag	Extracellular	20–30	[96]
Lactobacillus strains	Ag and Au	Intrecellular	-	[97]
Plectonema boryanum	Ag	Intrecellular	1–10	[98]
(Cyanobacteria)	CdS	Intracellular	2–5	[99]
Escherichia coli	Au	Intracellular	10–20	[100]
Shewanella algae	Au	Extracellular	10–20	[101]
Rhodopseudomonas	Au	Intracellular	25–33	[102]
capsulate	Au	Extracellular	8	[103]
Escherichia coli DH5α	Au	Intracellular	5–15	[104]
Thermomonospora sp.	Au	Extracellular	15–30	[105]
Rhodococcus sp.				
Pseudomonas aeruginosa				
(B) Yeast				
MKY3	Ag	Extracellular	2–5	[106]
Candida glabrata and *Schizosaccharomyces pombe*	CdS	Intracellular	200	[107]
(C) Fungi				
Phoma sp. 3.2883	Ag	Extracellular	71–74	[108]
Fusarium oxysporum	Au	Extracellular	20–40	[109]
Verticillium	Ag	Intracellular	25	[110]
Aspergillus fumigates	Ag	Extracellular	5–25	[111]
Trichoderma asperellum	Ag	Extracellular	13–18	[112]
(D) Algae				
Sargassum wightii	Au	Extracellular	8–12	[113]
Chlorella vulgaris	Au	-	9–20	[114]

delivery, and tumor targeting. The multifaceted applications of MNPs are detailed in the following sections.

Medical Diagnosis and Molecular Biology

The remarkable properties of metallic nanoparticles are harnessed for biomedical purposes. As particle size shrinks, their high surface area imparts distinct properties distinct from macro-sized structures. Metallic nanoparticle synthesis employs either bottom-up or top-down approaches. The bottom-up method, primarily through thermal decomposition, is cost-effective and flexible, and yields homogeneous particles. Notably, biological synthesis has gained prominence for its non-toxicity, cost-efficiency, sustainability, and eco-friendliness [116–118].

Bioimaging

Advancements in non-invasive techniques have revolutionized medical research, benefiting fields like drug development, diagnostics, and cellular biology. Innovations span MRI, electro-optic imaging, ultrasonic scans, positron emission tomography, and molecular bioimaging. Noble metals such as gold and silver nanoparticles play pivotal roles in enhancing imaging, capitalizing on their efficient photon emission in the near-infrared range. These nanoparticles serve as potent contrast agents [119–121].

Magnetic Resonance Imaging (MRI)

MRI, renowned for its superior spatial resolution, noninvasiveness, and non-destructiveness, has evolved due to nanotechnology. Nanoparticles (NPs) have unlocked possibilities for specialized therapeutic applications, particularly superparamagnetic iron oxide NPs, as contrast agents for cellular and molecular-level quality [122, 123].

Contrast Agents in X-ray Computed Tomography (CT)

X-ray computed tomography is pivotal in various medical applications but often requires contrast agents to improve visibility. Metallic nanoparticles, owing to their high X-ray absorption and specific gravity, are explored as alternatives to iodine-based agents. Efforts to reduce toxicity through surface modification with organic molecules are underway [124].

Tissue Construction

Nanoparticles have gained significance in tissue engineering, offering ideal properties such as low toxicity, adaptability, contrast-enhancing effects, active targeting, and precise control. Au nanoparticles and BGC nanoparticles (bioactive glass ceramic-based nanoparticles) are promising choices in stem cell therapy and tissue regeneration [125, 126].

Protein Detection

Proteins play pivotal roles in biological systems, necessitating their detection and understanding. Gold nanoparticles are frequently employed in immunocytochemistry for protein–protein interaction studies. Surface-enhanced Raman scattering spectroscopy enhances protein probe functionality [127, 128].

Drug Delivery

Metal nanomaterials offer promising avenues for efficient medical treatment and diagnosis. Their unique physicochemical properties, including high electrical and thermal stability, large surface area, and bioactivity, make them invaluable in drug delivery. Metal nanoparticles enhance drug permeability and retention, particularly through encapsulation, making them suitable for various therapies [129].

Cancer Drug Delivery

Effective cancer therapeutics strive for maximum bioavailability and site-specific action while minimizing harm to healthy tissues. Gold nanoparticles (AuNPs) exhibit low toxicity, ease of surface modification, and targeted activity on cancer cells. Silver nanoparticles (AgNPs) also display anticancer properties, inducing apoptosis and increasing ROS generation. Iron oxide nanoparticles (IONPs) offer high targeting capabilities under external magnetic fields, improving therapeutic efficacy with minimal damage to healthy cells. A range of metal nanoparticles, including zinc oxide (ZnO), copper oxide, cerium oxide (CeO), titanium dioxide (TiO2), and palladium nanoparticles (Pd NPs), are utilized for drug delivery in cancer treatment. These nanoparticles contribute to apoptosis, ROS production, and tumor suppression [130–132].

Nanovaccines

Metallic nanomaterials represent one of the most diverse classes of nanomaterials, characterized by a plethora of valuable properties. Numerous studies have shown that nanoparticles (NPs) possess the capability to stimulate immune responses, which encompass cell recruitment, activation of antigen-presenting cells (APCs), and the induction of cytokine and chemokine release. Scientific research has specifically demonstrated that metallic nanoparticles, such as silver nanoparticles, have the ability to activate immune cells like macrophages. Similarly, gold nanoparticles are known to induce cytokine secretion and exhibit efficient cellular penetration, thereby activating the first line of defense mechanisms through the stimulation of antigen-presenting cells [133, 134].

Cytotoxicity of Green Synthesized Metallic Nanoparticles

Main Consideration

A significant challenge in nanotechnology is the potential release of metal nanoparticles into the environment and their exposure to humans. The increasing commercial utilization of nanoparticles is likely to lead to elevated exposure levels, which could have adverse effects on human health. This concern falls within the extensive domain of nanotoxicology, which addresses these potential risks.

Various nanomaterials can enter the human body through ingestion via the gastrointestinal tract, absorption through the skin, or inhalation. The skin, comprising several protective layers, plays a vital role as a physiological barrier in both humans and animals. It serves as a defense mechanism against the spread of diseases or the harmful effects of substances. When nanoparticles are ingested, they are known to accumulate in the liver, an organ with a high metabolic rate, leading to issues such as glutathione depletion, impaired mitochondrial function, and increased oxidative stress [95]. Another area of concern for ingested nanoparticles is their potential impact on the respiratory system. Many metal nanoparticles have been found to adversely affect pulmonary function through oxidative mechanisms and other means [135, 136].

Cytotoxicity of Metallic Nanoparticles Manufactured by Green Synthesis

Nanomaterials, particularly those manufactured using environmentally friendly methods, hold significant promise in the medical industry, particularly as controlled drug delivery systems and potential anticancer agents [137]. However, there are still concerns about the safety of metallic nanoparticles used in anticancer treatments. Research has shown that the size of nanoparticles can negatively influence their cytotoxicity. Most metallic nanoparticles produced by plants are typically circular or nearly spherical, with a mean lethal dose of 120 g/mL. While less potent nanoparticle variants exist, certain plant-mediated manufacturing techniques have shown no harmful effects [138]. For instance, when assessing the cytotoxicity of Ag-NPs on the MOLT-4 cell line using the MTT assay, these nanoparticles exhibited remarkable antibacterial activity at low concentrations. According to cytotoxicity assays, environmentally produced Ag-NPs displayed an IC50 of 0.011 m, in contrast to 1.8 for cisplatin, a more potent monoclonal antibody for the MOLT-4 cell line [139].

Current Challenges Associated with Green Synthesized Metallic Nanoparticles

Challenges of Targeted Action

Effective therapeutic action necessitates the precise delivery of drugs from MNPs to the site of illness. Nanoparticles play a crucial role in assisting drug molecules in traversing various biological barriers. For instance, oral nanoparticles (NPs) must exhibit stability in gastric acid and navigate the intestinal epithelium to achieve systemic availability. Similarly, intravenously injected nanoparticles must surmount numerous biological obstacles to reach their designated targets. In the context of cancer therapy with MNPs, drug delivery is often hindered by the high density of cancer cells and the extensive tumor stroma [140, 141]. Additionally, the presence of high interstitial fluid pressure within tumors acts as a barrier, reducing transvascular drug delivery and resulting from impaired lymphatic drainage. To enhance efficiency and optimal internalization, the success of active targeting relies on the affinity and effectiveness of nanocarrier ligands [142].

Challenges of Green Synthesized Metallic Nanoparticles Analysis and Characterization

The complexity of metal nanoparticles necessitates advanced testing techniques to comprehensively describe the physical, chemical, and biological properties of created NPs. Despite the development of cutting-edge testing techniques for MNPs analysis, they often struggle to differentiate between active formulations and those that are inactive or less active [143].

Pharmacology and Safety Challenges

Conventional assessment of plasma drug concentration is a commonly used method to evaluate the pharmacokinetic properties of nanoparticles. However, due to the wide range of nano sizes exhibited by MNPs, achieving uniform pharmacokinetic properties is challenging, making it difficult to determine their bioequivalence and therapeutic efficacy. Furthermore, there is a scarcity of data regarding the in vivo toxicity of metal nanoparticles, particularly when administered over extended periods [144, 145].

Regulatory Challenges

While the FDA and the European Medicines Agency (EMA) have granted approval for several nanoparticles for use in cancer therapy, regulatory bodies have yet to establish guidelines for medicinal products involving soft matter. Consequently, the evaluation of NPs currently relies exclusively on time-consuming, specialized risk–benefit analyses for individual cases, leading to regulatory delays. To ensure the safe utilization of MNPs, there is an urgent need to develop state-of-the-art, versatile tools and establish comprehensive guidelines for nanoparticle applications [146, 147].

Summary and Future Perspectives

This comprehensive review delves into the world of metallic nanoparticles (MNPs), exploring their manufacturing techniques and their myriad of biological applications. Through meticulous examination of the existing literature and the practical deployment of nanosystems in clinical settings, this review provides a panoramic view of the recent developments in nanomedicine, with a particular focus on drug delivery systems. Over the past two decades, an array of innovative, cost-effective, and dependable synthesis methods has emerged. These advancements have not only enhanced the physicochemical properties of hybrid MNPs but have also unveiled novel possibilities in the realm of disease diagnosis and treatment. Recent research trends have ushered in a profound exploration of the diverse roles MNPs can play in the biological field. The versatility of MNPs has been greatly influenced by surface modifications and tailored functionalization, paving the way for their use in managing tissue and organ-related challenges, targeted drug delivery, singular-agent disease diagnostics and therapy, among other emerging healthcare applications.

MNPs have emerged as indispensable tools in the twenty-first century, making significant contributions to the fields of biology and medicine. Their pivotal role in enhancing drug targeting and delivery, especially in cancer treatment, has been widely acknowledged. Furthermore, MNPs have evolved to serve as effective imaging diagnostic tools for cancer cells, facilitating precise diagnosis and monitoring. In the biomedical arena, MNPs have proven to be a safe, efficient, and cost-effective alternative to conventional treatments. However, they encounter challenges when navigating biological barriers, limiting their potential impact. These biological constraints pose significant obstacles, potentially hindering pharmaceutical companies from investing in metallic nanomedicines. To overcome these barriers, it is imperative to deepen our understanding of disease pathophysiology and the inter-patient and intra-patient variations that exist. Modifying the physicochemical attributes of MNPs is another critical aspect to improve drug targeting towards the site of illness. The dearth of knowledge regarding the intricate interplay between patient biology and MNP behavior often results in optimistic expectations falling short in clinical studies. Addressing this issue requires a meticulous evaluation of preclinical data encompassing safety, effectiveness, bio-distribution, and pharmacokinetics, ideally in suitable animal models. While MNPs have shown promise as carriers for chemotherapeutic drugs through the enhanced permeability and retention (EPR) effect, its efficacy has been documented primarily in a limited number of tumor types due to the heterogeneous nature of tumors, inter-patient variability, and intra-patient variations. As such, a one-size-fits-all approach for nanoparticle size in cancer therapy should be avoided.

Synthesizing MNPs is a complex and challenging process, involving both chemical and green methods. The high manufacturing costs, intricacies of synthesis techniques, and the complex structures of MNPs present limitations for large-scale production. To harness the full potential of MNPs in the biomedical field, attention must be given to rethinking nanoparticle synthesis processes. While green synthesis offers a sustainable approach, it is imperative that the materials used are not only eco-friendly but also cost-effective and devoid of hazardous substances. Moreover, the clinical safety of MNPs requires thorough evaluation, given their potential for clinical toxicities. As various types of metal nanoparticles and composites gain popularity in the medical, biological, and healthcare sectors, comprehensive toxicological and safety assessments become increasingly essential. Strategies to mitigate the self-toxicity of MNPs include ion doping, functionalization, and the development of conjugated polymer-metal oxide nanoparticles.

Recent technological advancements have paved the way for extensive testing of MNPs for multifaceted applications. While their primary application today is the treatment of malignant cells, exploring MNPs as drug delivery systems (DDS) for various therapeutic drug classes is gaining momentum. Additionally, MNPs' antibacterial properties could be harnessed to create medical devices with self-disinfection capabilities, potentially offering a cost-effective alternative to conventional antibiotics.

In conclusion, addressing the challenges associated with MNPs holds the promise of significantly advancing their clinical translation for various biological applications, including cancer treatment and beyond. The ongoing quest for solutions will undoubtedly contribute to the evolving landscape of MNPs in healthcare [148, 149].

Acknowledgements The authors are thankful to the Faculty of Pharmacy, Integral University, Lucknow, for providing all the necessary facilities related to the present work (Manuscript Communication Number: IU/R&D/2022-MCN0001416).

Author Contribution P.K. conceived and designed the study, and wrote the first draft of the manuscript, S.S. carried out the research, B.S. and M.A participated in the analysis and interpretation of data. All authors contributed to and approved the final draft of the manuscript.

Data Availability The data that support the findings of this study are available upon reasonable request.

Declarations

Ethics Approval No human or animal subjects were used in the course of the research for this study.

Competing Interests The authors declare no competing interests.

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