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Research Paper

Natural product-based nanoparticles: phytochemical insights, bioactivity profiling, and biomedical applications

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ABSTRACT

PDNPs serve as a bridge between natural product chemistry and nanomedicine. It is their unique physical and chemical properties that render PDNPs capable of targeting intractable infectious diseases, inflammation, cancer, and drug-resistant pathogens. This will dedicate most of the efforts to summarizing recent publications and researches on the extraction methods and bioactivity, and production of PDNPs and their applications in biomedicine. Polyphenols, flavonoids, alkaloids and terpenoids rich plant extracts serve both as a reducing and capping agent epitomized in most cases to nanoparticle synthesis utilizing green methods. The biosynthesized nanoparticles fabricated in this manner, display significant antimicrobial, anti-inflammatory, anticancer, and immune-modulatory prowess. Key characterization techniques with emphasis on UV-Vis spectroscopy, FTIR, XRD, SEM/TEM, and DLS techniques and their relevance to evaluation of the nanoparticles for stability, shape, and size distribution are outlined. The bioactivity of PDNPs elicits positive responses on resistant pathogens and inflammatory mediators. This is evident in both the in vivo and in vitro studies. Investigational PDNP studies for Inflammatory Bowel Disease (IBD), wound healing, drug delivery, and cancer diagnostics and therapeutics have been met with mounting enthusiasm. The new role of plant-derived exosome-like nanovesicles and vesicle-like nanoparticles in precision medicine is also discussed. Standardization, toxicology studies, and feasible research are emphasized based on data from over 35 recent research studies. Targeted delivery, regulatory harmonization, and sustainable development in nano-phytomedicine are among the future directions.

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1. INTRODUCTION

The intersection of nanotechnology and phytochemistry has ushered in a new era in biomedical innovation with the development of plant-derived nanoparticles (PDNPs) [1]. These nanoscale entities, made from plant extracts, hold great potential due to their compatibility with biological systems, environmental sustainability, and ability to target multiple biological processes [2]. Unlike traditional synthetic nanoparticles, which often rely on toxic chemicals, PDNPs utilise phytochemicals as natural reducing and stabilising agents [3]. This results in environmentally friendly and cost-effective production methods [4]. In the past, medicinal plants have been a treasure trove of drugs because of their broad range of bioactive secondary metabolites such as flavonoids, alkaloids, saponins, and tannins [5]. Modern developments have focused attention from bulk phytochemicals towards nanosized formulations. Such formulations can add therapeutic value by virtue of increased bioavailability, targeting, and controlled release [6]. These developments are significant in unraveling complex health problems like antibiotic resistance, chronic inflammation, cancer, and inflammatory bowel disease [7].

The idea of phyto-nanotechnology, where plant molecules are used to develop nanoparticles, has transformed our understanding of natural product delivery systems [8]. Such biogenic nanoparticles have a wide range of antimicrobial, anti-inflammatory, anticancer, and immunomodulatory functions and often work better and more safely than conventional drugs [3], [9]. In addition, plant-derived exosome-like nanovesicles have emerged as the new targeting tool [10]. Since nanoparticulate PDs are natural vesicle-forming units that can cross biological barriers and transport drugs without eliciting an undesirable immune reaction, they have very useful properties in drug delivery [11]. Research studies have shown them to be capable of treating inflammatory diseases and cancers as well as neurological disorders [12].

This article is meant as a review of recent advancements in the PDNPs field, focusing on their synthesis, characterization, and application in modern medicine. The formation process involves appropriate extraction and phytochemical profiling of the plant material with various bioactive constituents such as flavonoids, alkaloids, and terpenoids. Such phytochemicals are a necessity, for they act as destructive and stabilizing substances in the green synthesis routes later used for nanoparticle formation. The green synthesis route, which avoids the use of toxic chemicals, guarantees biocompatible and environmentally friendly nanoparticles.

To establish nanoparticle properties, diverse tools for structural and morphological characterizations are engaged. UV-Vis spectroscopy validates nanoparticle formation and offers optical properties. Microscopy techniques include cultivating size and shape images using Scanning Electron Microscopy (SEM) and Transmission Electron Microscopy (TEM), and characterizing the crystalline structure of these particles using X-ray Diffraction (XRD). The review also speaks of the versatile biomedical uses of PDNPs, which take advantage of their exclusive properties. These involve strong antimicrobial activity against a wide range of pathogens, strong anti-inflammatory activity by suppressing oxidative stress, and potential anticancer activity through targeted drug delivery and induction of apoptosis. Synergism between the plant's bioactive compounds and the properties of the nanoparticle makes PDNPs a very effective and promising field in nanomedicine.

2. RELATED WORK

Various plant materials like medicinal plants, food plants, leaf extracts, and polyherbal sources are utilized to prepare various types of nanoparticles like metal NPs, plant-derived nanoparticles (PDNPs), plant-virus nanoparticles (PVNPs), and polyherbal nanoparticles (PdNPs) [13]. One major trend is the

widespread exploitation of these nanoparticles for their antimicrobial and anti-inflammatory activities, and use in wound healing and infection control. The table also reflects their prospect in higher-end applications such as cancer therapy, delivery of nutraceuticals, and immune system modulation for the treatment of diseases such as Inflammatory Bowel Disease (IBD) [14], [15].

Usually, nanoparticles need to be characterized by a combination of techniques. UV-Vis absorption spectroscopy verifies the formation of nanoparticles, while the FTIR spectrum informs about the functional groups on the plant material that may be responsible for synthesis and stabilization [16]. SEM and TEM have been used for characterizing morphology and size. Dynamic light scattering (DLS) and nanoparticle tracking analysis (NTA) are used for measuring size distribution. The method of studying the crystalline nature is X-ray diffraction (XRD), while liquid chromatography-mass spectrometry (LC-MS) discloses the chemical composition of the plant extracts [17], [18]. Table 1 offers a comparison of plant-based nanomaterials (NPs) from various natural resources with emphasis on their diversity, key bioactivities, and common characterization techniques as well as applications.

Table 1. Comparative Study of Plant Materials

Plant/Source	Type of NP	Key Bioactivity	Characterization Methods	Application
Various medicinal plants	Metal NPs	Regenerative, Antimicrobial	UV, FTIR, SEM, TEM	Wound healing
Edible plants & viruses	PDNPs & PVNPs	Drug delivery, Immune modulation	NTA, DLS	Nutraceutical delivery
Leaf extracts	AgNPs, ZnNPs	Antimicrobial, Anti-inflammatory	UV, XRD, FTIR	Infection control
Nanovesicles	ELNs	Immune modulation	TEM, NTA, LC-MS	IBD, Cancer
Polyherbal sources	PdNPs	AMR suppression	GC-MS, DLS	Antibacterial
Polyphenol-rich plants	AuNPs, AgNPs	Drug delivery	XRD, UV, FTIR	Cancer therapy
Citrus & cruciferous vegetables	ELNs	Anti-inflammatory	Zeta, LC-MS	IBD

3. METHODOLGY

Preparation and application of plant-derived nanoparticles (PDNPs) is yet another science discipline. It is a marriage of traditional plant extraction with present-day nanotechnology and testing methods. The process has many significant steps, ranging from plant preparation to the extraction of its chemicals, nanoparticle preparation, structural analysis of the nanoparticles, and activity testing of the nanoparticles. All the steps are significant to make the nanoparticles stable, active, and potent for biomedical applications.

A. Plant Extract Preparation

The process starts with collecting and preparing plant materials. This usually involves drying and grinding leaves, stems, roots, or fruits, depending on the plant's chemical makeup [19], [20]. Drying often happens in the shade to protect sensitive compounds. At this stage, the plant material is roughly ground using a mechanical grinder to increase surface area, which results in better interaction with solvents [21]. Once it is powdered, the plant material is ready for extraction by using either organic or water-based solvents. Common solvents include methanol, ethanol, and distilled water. These solvents efficiently extract phenolic compounds, flavonoids, and terpenoids [22].

Soxhlet extraction is a common technique. It provides continuous flow of the solvent over the plant material so that the chemicals are extracted properly [23]. The extract is then subjected to rotary evaporation under controlled temperature and reduced pressure. This drives off the solvent without degrading the phytochemicals [24]. Freeze-drying or lyophilization is applied in some researches,

particularly those where there are heat-sensitive compounds, in order to concentrate the plant extract without altering its chemical composition [2], [3]. The concentrated final extract serves as the reducing and capping agent in the preparation of nanoparticles in subsequent steps [25].

B. Phytochemical Analysis Extract Preparation

Before the process of synthesis, the extract is screened for its phytochemical composition, as availability and concentration of certain metabolites such as flavonoids, alkaloids, saponins, and polyphenols can influence the formation and stability of the nanoparticles [23]. Phytochemical qualitative screening entails routine colorimetric assays for general identification of classes of compounds: e.g., Mayer's reagent for the detection of alkaloids, ferric chloride for phenolic compound presence, and Shinoda test for flavonoids. Quantitative analysis is then followed by the measurement of the total phenolic content (TPC), total flavonoid content (TFC), and other bioactive compounds through methods such as High-Performance Liquid Chromatography (HPLC), Liquid Chromatography-Mass Spectrometry (LC-MS), and UV-visible spectrophotometry [24]. These techniques not only validate the bioactive abundance of the extract but also assist in batch standardization for reproducibility [14]. These types of profiling are crucial because differences in phytochemicals due to species, geospatial location, or season may alter the size, shape, and stability of nanoparticles synthesized [25].

C. Nanoparticle Synthesis

This green synthesis is regarded to be the most crucial step in PDNP fabrication. This process is preferred above physical and chemical processes due to its simplicity, toxicity-free nature, and eco-friendliness. During this synthesis, a metal salt solution, typically silver nitrate ($AgNO_3$), zinc sulfate ($ZnSO_4$), or copper sulfate ($CuSO_4$), is mixed with plant extract in a certain volume ratio [25]. Under controlled pH, temperature, and stirring conditions [26], the above occurs. Bioactive compounds present in the extract, like phenolics and flavonoids, reduce metal ions to their zero-valent metallic state. These bioactive compounds are also capping agents to avoid aggregation and enhance stability [27].

A colour change is usually visible to initiate the synthesis of nanoparticles. As an example, yellowish-brown in AgNPs can be observed because of the Surface Plasmon Resonance (SPR) effect. This transformation is monitored and verified by UV-Vis spectroscopy, where a clear peak of absorbance typically appears between 350 nm to 450 nm for AgNPs and ZnNPs. Size, shape, and variation functionalities that are attributed to metal ion concentration, reaction time, temperature, and pH of the synthesis process [28].

D. Characterization Techniques

After synthesis, PDNPs undergo thorough physicochemical characterization determining structure, morphology, and surface chemistry. The first verification is done through UV-visible spectrophotometry detecting SPR peaks specific to the particular nanoparticles [29]. Fourier Transform Infrared Spectroscopy (FTIR) then detects the functional groups present on the surface of the nanoparticle, indicating the presence of phytochemicals as capping agents [5], [30].

X-ray Diffraction (XRD) analysis helps establish the crystalline nature and phase of the nanoparticles. Common diffraction patterns show face-centred cubic structures for silver and zinc-based nanoparticles. Scanning Electron Microscopy (SEM) and Transmission Electron Microscopy (TEM) offer high-resolution images that reveal size distribution, surface shape, and form, such as spherical, triangular, or rod-like. Most PDNPs synthesized range in size from 10 to 100 nm, which is ideal for cellular uptake.

Furthermore, Dynamic Light Scattering (DLS) establishes the hydrodynamic diameter and polydispersity index (PDI) of nanoparticles in colloidal solution, through which stability can be understood. Analysis of Zeta potential is also insightful in terms of surface charge, and over ±30 mV values indicate good stability and anti-aggregation. These characterization processes prove to be highly important for maintaining the consistency, efficacy, and safety of PDNPs in biomedical applications.

E. Bioactivity Assay

After the nanoparticles have undergone successful synthesis and characterization, they are subjected to a battery of bioactivity tests against the putative therapeutic targets to confirm their biological activities. For antimicrobial activity, the classical disc diffusion method on agar plates and MIC assays are carried out. The tests measure the diameter of the inhibition zone and the minimal concentration inhibiting visible growth, respectively. Some researchers have found that PDNPs produce more antimicrobial activity than that of standard phytochemical extraction due to being able to penetrate better and to generate reactive oxygen species. For its anti-inflammatory activity, protein denaturation inhibition, membrane stabilization, and nitric oxide (NO) inhibition in macrophage cell lines are routine tests. The ability of nanoparticles to inhibit protein denaturation is similar to the potential of nanoparticles in arresting inflammation-induced diseases. NO inhibition, as measured using the Griess assay, corresponds to the inhibition of the production of pro-inflammatory cytokines, a pivotal process in autoimmunity and chronic inflammation. The MTT assay, a colorimetric assay, is among the most widely used techniques for anticancer or cytotoxic activity estimation. It works on the principle of measuring mitochondrial activity as an indirect measure of cell viability. Nanoparticles exhibiting dose-dependent cytotoxicity towards cancer cell lines, e.g., MCF-7 and HeLa, are also investigated for mechanisms such as apoptosis induction and cell cycle arrest. Other techniques, such as trypan blue exclusion and flow cytometry, assist in establishing these findings.

4. RESULTS AND DISCUSSION

Drug delivery has the highest number of studies and almost equal in antimicrobial and anti-inflammation application. Other significant areas include cancer therapy, diagnostics, and IBD treatment, highlighting the diverse potential of plant-derived nanoparticles, as shown in Figure 1.

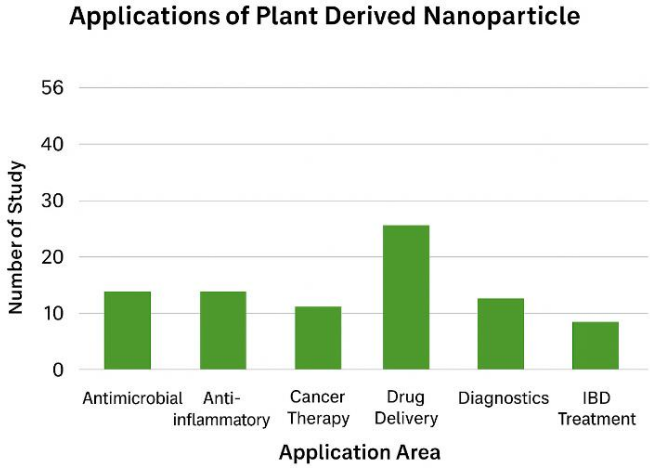

Figure 1. Applications of Plant-Derived Nanoparticles

A. Overview of Biological Applications of PDNPs

Applications of plant-derived nanoparticles (PDNPs) include a broad range of biomedicine fields. The reviewed literature and illustrated in the accompanying bar chart, the most significant research fields are drug delivery and antimicrobial properties. This is followed by anti-inflammatory, anticancer, and theranostic applications. The distribution illustrates the wide scope of therapeutic measures PDNPs require. A phytochemical can be encapsulated within a nanoparticle that offers targeted delivery into particular parts and thus greatly increase the therapeutic effects of many plant-based drugs.

B. Size-Dependent Bioactivity of PDNPs

Size remains an important factor that may affect biological activity of PDNPs. It is to be emphasized that scientists have shown repeatedly that nanoparticles grant enormous cellular internalization when their diameter ranges below 100 nm. This is because they possess a better surface area-to-volume ratio and can more readily pass through biological barriers. Smaller particles can interact with microbial membranes more efficiently, penetrate tumor tissue more effectively, and escape rapid elimination from the immune system. For instance, silver nanoparticles (AgNPs) synthesized from Azadirachta indica exhibited maximum antimicrobial efficacy when their diameter was maintained between 20 and 80 nm. This size-dependent characteristic plays a very important role in the design of nanoparticles for targeted therapy.

C. Surface Charge and Stability: Role of Zeta Potential

Zeta potential, an indicator of the surface charge of nanoparticles, decides their colloidal stability. Experiments suggest that nanoparticles with a zeta potential greater than ±30 mV generally will be dispersed and agglomeration resistant under physiological conditions [14]. Enhanced stability means extended shelf life, ensures constancy in biomedical assays, and facilitates distribution in the body. Most importantly, stable zeta potential can impact PDNPs' cell membrane interaction, improving their therapeutic index.

D. Multi-Functional Biological Activities

They are multifunctional in nature because PDNPs find themselves acting on several fronts simultaneously. It shows antimicrobial and anti-inflammatory and anticancer activities, all in one formulation. This biological plurality is chiefly due to complex compounds found in extracts of plants since many compounds exhibit synergistic effects. For example, silver nanoparticles synthesized using Ocimum sanctum extracts have shown strong antimicrobial activity against Escherichia coli and Staphylococcus aureus. They also reduced pro-inflammatory cytokines in lipopolysaccharide (LPS)-stimulated macrophages [11], [27]. This dual action makes PDNPs especially useful for treating infections where inflammation is also a concern.

E. Anticancer Potential of Phytogenic Nanoparticles

Several studies have looked at using PDNPs in cancer therapy. They are known for their ability to cause apoptosis and reduce tumor growth. The gold nanoparticles (AuNPs) synthesized using Curcuma longa and Azadirachta indica extracts have shown pronounced cytotoxicity towards MCF-7 breast cancer cells. They induce cell death by causing mitochondrial dysfunction and activating caspases [9], [18]. These formulations often show selective toxicity, targeting cancer cells more than healthy ones. This, therefore, lowers the side effects while representing an improvement over traditional chemo.

F. PDNPs in Managing Inflammation

Here exists yet one more example in which these PDNPs have shown immense potential. For example, ginger nanoparticles have been shown to demonstrate the anti-inflammatory property in cell and animal models of Inflammatory Bowel Disease (IBD). The particles further reduced the expressions of key proinflammatory cytokines (TNF-α, IL-6) and of mucosal repair genes in DSS-induced colitis mice. The same way, [7] discovered that nanoparticles from broccoli not only inhibited colon inflammation but also delayed the progression of cancer induced by colitis. This suggests their therapeutic and preventive value [3], [4].

G. Drug Delivery and Targeted Therapeutics

One of the most studied and successful uses of PDNPs is in drug delivery. Their ability to encapsulate hydrophobic drugs, protect them from breakdown, and deliver them directly to target tissues has opened new paths in targeted nanomedicine. Research shows that plant-derived exosome-like nanoparticles from grapefruit, ginseng, and turmeric can effectively transport drugs across biological barriers like the intestinal epithelium and the blood-brain barrier [18], [20], [24], [27]. They serve as the

most natural carriers, are completely biodegradable, and non-toxic. They also assist in increasing oral absorption of many poorly soluble drugs.

H. Biocompatibility and Selectivity

An outstanding feature of PDNPs, as opposed to synthetic nanoparticles, is thereby their high biocompatibility. The synthesis involves the use of natural capping agents, and the active ingredients are derived from edible or medicinal plants. This reduces the cytotoxicity risk, immunogenicity, or tissue accumulation immensely [7]. Some studies in the literature have also provided evidence that PDNPs exhibit a very selective mechanism, especially in the treatment of cancer. These are reported to be selectively cytotoxic against cancerous cells, and they have no adverse effect on normal cells, an attribute that most conventional chemotherapeutics lack [24], [27].

I. Limitations and Challenges

Despite their potential, the translation of PDNPs from the bench to the bedside faces many challenges. Variability in plant material, caused by differences in species, geography, or season, can lead to inconsistency in nanoparticle composition and activity. Additionally, the absence of standardized protocols for extraction, synthesis, and characterization makes it hard to compare findings across studies. Limited in vivo toxicology data and regulatory challenges also hinder clinical adoption. Facing such issues will be forefront to the industrial scale production and approval processes of PDNP-based therapeutic agents.

5. CONCLUSION

Plant-derived nanoparticles mark an essential chapter in nanomedicine history, entirely in opposition to those produced by synthetic methods in being more versatile and biocompatible. These nanoparticles carry broad therapeutic bioactivities, such as antimicrobial, anti-inflammatory, and targeted antitumor therapies, thus indicating a large therapeutic potential. However, several serious issues must be tackled before their evolution from laboratory life to practical existence. Priorities for the future must address standardization of synthesis and extraction procedures, such that batch-to-batch consistency and reproducibility are ensured. Also, comprehensive toxicity profiling with in vivo models is needed to ascertain safety. Further, their clinical utility needs validation, and industrial scale production methods for fabrication have to be developed, particularly dependable methods. In this way, the present work will be bridged with the prospect of plant-based nanoparticles entering extensive clinical use.

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Name of Author	C	M	So	Va	Fo	I	R	D	O	E	Vi	Su	P	Fu
Apurwa Singh	✓				✓	✓		✓	✓				✓	
Parinita Tripathy	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓	✓		

C : **C**onceptualization
M : **M**ethodology
So : **So**ftware
Va : **Va**lidation
Fo : **Fo**rmal analysis
I : **I**nvestigation
R : **R**esources
D : **D**ata Curation
O : Writing - **O**riginal Draft
E : Writing - Review & **E**diting
Vi : **Vi**sualization
Su : **Su**pervision
P : **P**roject administration
Fu : **Fu**nding acquisition

Conflict of Interest Statement

No conflict of interest.

Informed Consent

We have obtained informed consent from all individuals included in this study.

Ethical Approval

Not applicable.

Data Availability

Data availability does not apply to this paper as no new data were created or analyzed in this study.

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