

Polymeric nanoparticles for protein and peptide delivery

Nijhawan Monika, Neerude Sirisha, Nawale Sneha*

Department of Pharmacognosy, Gokaraju Rangaraju College of Pharmacy, Hyderabad 500090, India

* Corresponding author: Sneha Nawale, sneha.nawale11@gmail.com

CITATION

Monika N, Sirisha N, Sneha N. (2024). Polymeric nanoparticles for protein and peptide delivery. *Characterization and Application of Nanomaterials*. 8(1): 8291. <https://doi.org/10.24294/can8291>

ARTICLE INFO

Received: 30 July 2024

Accepted: 7 November 2024

Available online: 2 December 2024

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Abstract: Protein- and peptide-based medications are recognized for their effectiveness and lower toxicity compared to chemical-based drugs, making them promising therapeutic agents. However, their application has been limited by numerous delivery challenges. Polymeric nanostructures have emerged as effective tools for protein delivery due to their versatility and customizability. Polymers' inherent adaptability makes them ideal for meeting the specific demands of protein-delivery systems. Various strategies have been employed, such as enzyme inhibitors, absorption enhancers, mucoadhesive polymers, and chemical modifications of proteins or peptides. This study explores the hurdles associated with protein and peptide transport, the use of polymeric nanocarriers (both natural and synthetic) to overcome these challenges, and the techniques for fabricating and characterizing nanoparticles.

Keywords: drug delivery; nanoparticles (NPs); protein delivery; therapeutic effect

1. Introduction

Protein is derived from the Greek word "PROTOS", meaning the first or the supreme. Proteins are large organic molecules composed of amino acids linked together in a linear chain by peptide bonds, with proteins typically containing more than 50 amino acids. Peptides are short polymers formed by the linkage of amino acids in a specific order (a peptide contains < 50 amino acids). Proteins exhibit various structural forms: the primary structure, which is the sequence of amino acids; the secondary structure, consisting of regularly repeating local structures stabilized by hydrogen bonds; the tertiary structure, representing the three-dimensional configuration of the polypeptide; and the quaternary structure, formed by the assembly of multiple protein molecules (polypeptide chains) [1]. Proteins and peptides can be classified into different types, including polypeptides, oligopeptides, fibrous proteins, globular proteins, and oligomeric proteins, based on the number of amino acids present.

Proteins are essential to the body as they participate in various biological roles in the form of enzymes that catalyze virtually all chemical reactions (e.g., 6GDH), help in the transport of hemoglobin of erythrocytes, contract muscles (actin and myosin), maintain the structure of the body (collagen in bones), provide defensive activity (immunoglobulins and antibodies), regulate secretions (insulin), provide nutrition and storage in the body (ovalbumin), generate and transmit nerve impulses, provide immune protection through antibodies, and control growth.

Bioapplications of proteins and peptides include the use of erythropoietin to stimulate red blood cell production, tissue plasminogen activator for treating heart attacks and strokes, oxytocin to manage labor pain, bradykinins to enhance peripheral circulation, somatostatin to reduce bleeding in gastric ulcers, gonadotropin to induce

ovulation, and insulin to regulate blood sugar levels. Although proteins and peptides offer many benefits, their delivery poses challenges due to their large size and instability. Their structures are maintained by weak noncovalent forces, making them susceptible to degradation under mild storage conditions and gastric juices [2–4].

2. Problems with protein and peptide

The primary challenges in delivering proteins and peptides are proteolysis by *exo/endo* proteases, small-size proteins getting filtered out by kidneys very easily, causing elimination of B and T cells, and may show unwanted allergic reactions (even toxicity), and less therapeutic activity of proteins due to insolubility/adsorption [5].

3. Barriers to protein and peptide delivery

3.1. Enzymatic barriers

Proteins and peptides can be degraded by enzymes in two distinct ways. One method involves the hydrolytic breaking of peptide bonds by enzymes that degrade insulin, convert angiotensin, and renin. Since proteolysis is an irreversible event, it may harm medications that include proteins and peptides. Others include chemical modification of proteins by oxidizing them using glucose oxidase or xanthine oxidase or phosphorylating them with kinases [6].

3.2. Intestinal epithelial barrier

It prevents protein medications from passing through the intestinal epithelium. Transport of protein and peptide medicines across the intestinal epithelium is accomplished by several processes, including paracellular movements, endocytosis, transcytosis, passive transport, and carrier-mediated transport. Dipeptides and tripeptides from the small intestine are extensively absorbed through active transport. Proteins and peptides that are too large to be absorbed through carrier-mediated transport can be taken up via endocytosis, which is one of the processes by which cellular internalization of proteins and peptides occurs that include pinocytosis (cell drinking) and phagocytosis (cell eating). Paracellular migration and persorption are the two mechanisms involved in drug absorption. The epithelial mucosa of the small intestine serves as a barrier against macromolecule penetration [7].

3.3. Capillary endothelial barrier

Proteins and peptides must either cross the endothelial cells themselves or move between the cells to pass through the capillary endothelium. Cytoplasmic enzymes can alter or metabolize solutes that pass through endothelial cell membranes. Therefore, the endothelial passage presents an enzymatic or metabolic barrier to the passage of the proteins and peptides [8].

3.4. Blood-brain barrier

One of the main barriers to protein delivery to the brain compartment is the blood-brain barrier (BBB). It is made up of the blood-cerebrospinal fluid barrier and the vascular BBB. BBB is made up of a monolayer of cells at both locations that are joined

by tight junctions and contain additional mechanisms to prevent or slow the flow of plasma into the central nervous system, permitting only the passage of uncharged, tiny, lipophilic molecules and gases. Proteins and other large molecules cannot cross the BBB [9].

Different barriers to protein and peptide drug delivery are illustrated in a visual format in **Figure 1**.

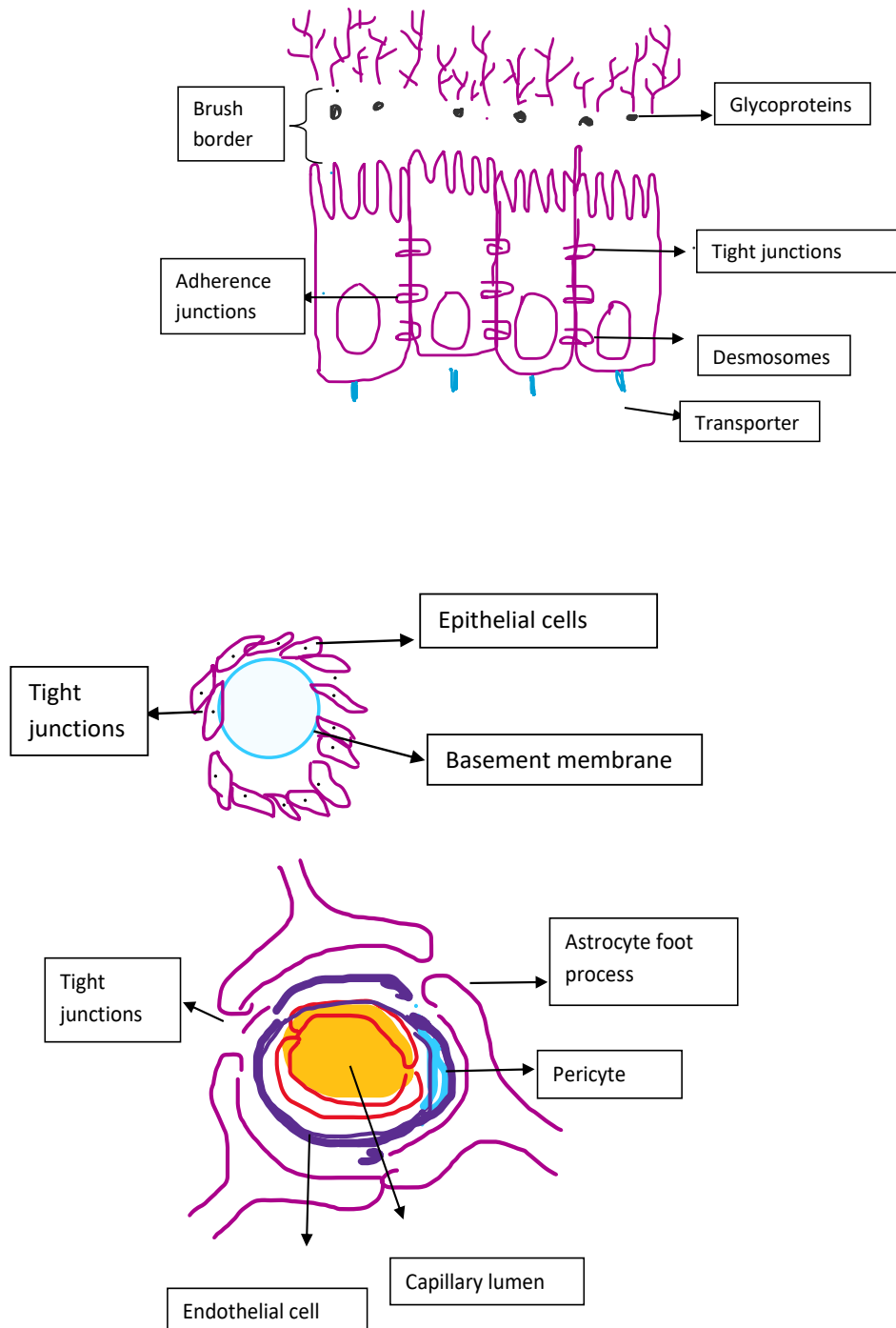


Figure 1. Intestinal barrier, capillary endothelial barrier and blood brain barrier for protein and peptide delivery.

4. Polymeric nanocarriers for protein and peptide delivery

Polymeric nanoparticles (NPs), which are solid colloidal carriers ranging from 10 to 100 nm in size, can be formulated with synthetic, semi-synthetic, or natural polymers. The choice of materials used to synthesize these NPs affects their drug delivery performance and therapeutic effects. Proteins can be chemically attached, adsorbed, or encapsulated on the surface of polymeric NPs [10,11].

4.1. Natural polymers

Therapeutic proteins can be delivered to specific sites for distinctive benefits with natural polymers. The natural polymers may be of plant, animal, and marine origin and are generally inexpensive. The existence of reactive sites in natural polymers is significant in drug delivery systems because it facilitates ligand conjugation, cross-linking, and other modifications that make the polymers perfect drug carriers for a variety of therapeutic proteins. However, natural polymers are more susceptible to processing parameters.

4.1.1. Polysaccharide-based natural polymers

Polysaccharides exhibit structural and functional diversity because of their vast number of reactive groups, wide range of molecular weights, and varied chemical composition. Being naturally occurring biomaterials, polysaccharides are safe, nontoxic, and biodegradable and exhibit remarkable stability in biological fluids. Polysaccharides are also known as mucoadhesive polymers because of their mucoadhesive qualities (polysaccharides, which are hydrophilic in nature, create non-covalent connections with biological tissues) due to the presence of many derivable groups on the molecular structure, such as hydroxyl, carboxyl, and amino groups. Mucoadhesive polysaccharide-based NPs have been demonstrated to improve the residence and absorbance time of integrated therapeutic protein. Various polysaccharides used for the transport of proteins and peptides are represented in **Table 1**.

Table 1. List of various polysaccharide polymers for proteins and peptide drug delivery.

Polysaccharide polymers	Properties	Role of polymers in transportation of Protein and Peptides	Reference
Chitosan	Biocompatible, Non-toxic	Aid in facilitating the intestinal epithelial mucosal absorption of therapeutic proteins with high molecular weight.	[12,13]
Cyclodextrins	Utilized as potential carriers in pharmaceutical biotechnology and non-toxic	Improve the bioavailability of the protein molecule by delivering it directly to the barrier membrane	[14,15]
Alginates	Excellent mucoadhesive qualities, Biocompatibility, biodegradability, High degree of flexibility and non-toxic	Helps in transport of therapeutic proteins that are heat sensitive. Alginate also transports labile proteins and peptides safely to the colon while shielding them from the stomach environment.	[16,17]
Pectins	Mucoadhesive activity Inert for physiological fluids and non-toxic	Stops integrated proteins from being broken down by gastric enzymes and greatly boosts the intestinal absorption by its unique mucoadhesive activity on the intestinal epithelium.	[18,19]
Xanthan Gum	Biocompatible, Non-toxic, biodegradable and bioadhesive properties	Preserves the integrity of the therapeutic protein and extends its sustained release	[20,21]

4.1.2. Protein-based polymers

Amidst natural polymers, protein-based polymers have garnered significant attention because of their attributes such as abundance, accessibility, minimal toxicity, modifiability due to their intricate heterogeneity, and adaptability in delivery methods as depicted in **Table 2**. There is still more work to be done to make protein-based polymers stable enough to be used as the perfect therapeutic polymers.

Table 2. List of various protein polymers for proteins and peptide drug delivery.

Protein polymers	Properties	Role of polymers in transportation of Protein and Peptides	Reference
Lectins	Sugar-binding proteins	Ability to improve the active transport of therapeutic proteins with large molecular weights from the intestinal epithelium	[22]
Albumin	Non-toxic, Biocompatible	Deliver therapeutic proteins across the blood-brain barrier and nuclear membrane	[23]
Collagen	Biocompatible Easily modifiable and available Synergic with bioactive components	Facilitates the delivery of therapeutic proteins and extends the sustained release of the incorporated proteins.	[24]
Gelatin	Biocompatible Biodegradable	Thermo-reversible properties are utilized for delivering therapeutic molecules through targeted drug delivery systems	[25]

4.2. Synthetic polymers

Formulators are focusing on synthetic polymers for the delivery of therapeutic proteins and peptides. It has been demonstrated that these polymers lengthen the pharmacokinetic and circulation periods of integrated medicinal compounds. Drug carriers made of synthetic polymers frequently serve a passive purpose as tabulated in **Tables 3 and 4**.

Table 3. List of various synthetic biodegradable polymers with their properties and transport of proteins and peptide drug delivery.

Synthetic polymers	Properties	Role in transportation of Protein and Peptides	Reference
Polyethylene Glycol	Non-immunogenic, Nontoxic, Highly soluble in water	PEGylation of therapeutic proteins protects them from enzymatic degradation and reduces immunogenicity, thereby extending their residence time in the body, enhancing stability, and modifying pharmacokinetics by altering various physicochemical properties	[26,27]
Polaxomers	Thermosensitive Inert and stable	Helps to sustain the stability of incorporated therapeutic proteins and peptides more effectively than other prolonged release drug delivery systems	[28,29]
Poly (lactic-co-glycolic acid)	Biocompatible and Biodegradable	Several types of PLGA-PEG block copolymers have been developed for the proteins and peptides sustained delivery	[30,31]
Pluronic F127	Non-irritant Biocompatibility Good mechanical strength Bioadhesive properties	Ensures greater stability of incorporated therapeutic proteins and peptides compared to other prolonged release drug delivery systems	[32]

Table 4. List of various synthetic non-biodegradable polymers with their properties and transport of proteins and peptide drug delivery.

Synthetic polymers	Properties	Role in transportation of Protein and Peptides	Reference
Silicons	High loading capacity, surface functionalization, biocompatibility, physicochemical and thermal stability	Protection of proteins from degradation, increase the half-life of protein, targetted delivery, ability to functionalize their surfaces	[33]
Polyacrylate	Versatile, non-toxic, mucoadhesive	Safe delivery of protein, controlled and sustained release of proteins and peptides	[34]

4.2.1. Synthetic biodegradable polymers

Biodegradable polymers offer significant advantages in biomedical applications, especially for drug delivery. Their capacity to break down into smaller, absorbable molecules eliminates the need for surgical removal, improving patient comfort and compliance. Moreover, their biocompatibility and non-toxicity make them ideal for use in a range of medical devices and treatments, reducing the risk of adverse reactions and promoting better integration within the body. This makes them a favored option for many cutting-edge therapeutic solutions [35,36].

4.2.2. Synthetic non-biodegradable polymers

Non-biodegradable polymers present considerable challenges in medical applications, as they require surgical removal once the medication, they deliver is exhausted. This limitation confines their use to cases where the implant can be easily retrieved. On the other hand, biodegradable polymers naturally break down within the body, providing a more convenient and less invasive alternative for drug delivery systems. This distinction highlights the importance of choosing the appropriate polymer type based on the specific medical requirements and desired treatment outcomes.

5. Method of preparation of nanocarriers

Typically, two primary approaches are utilized, which are the dispersion of prefabricated polymers or the polymerization of monomers. The lists of commonly used techniques are presented in **Figures 2** and **3**.

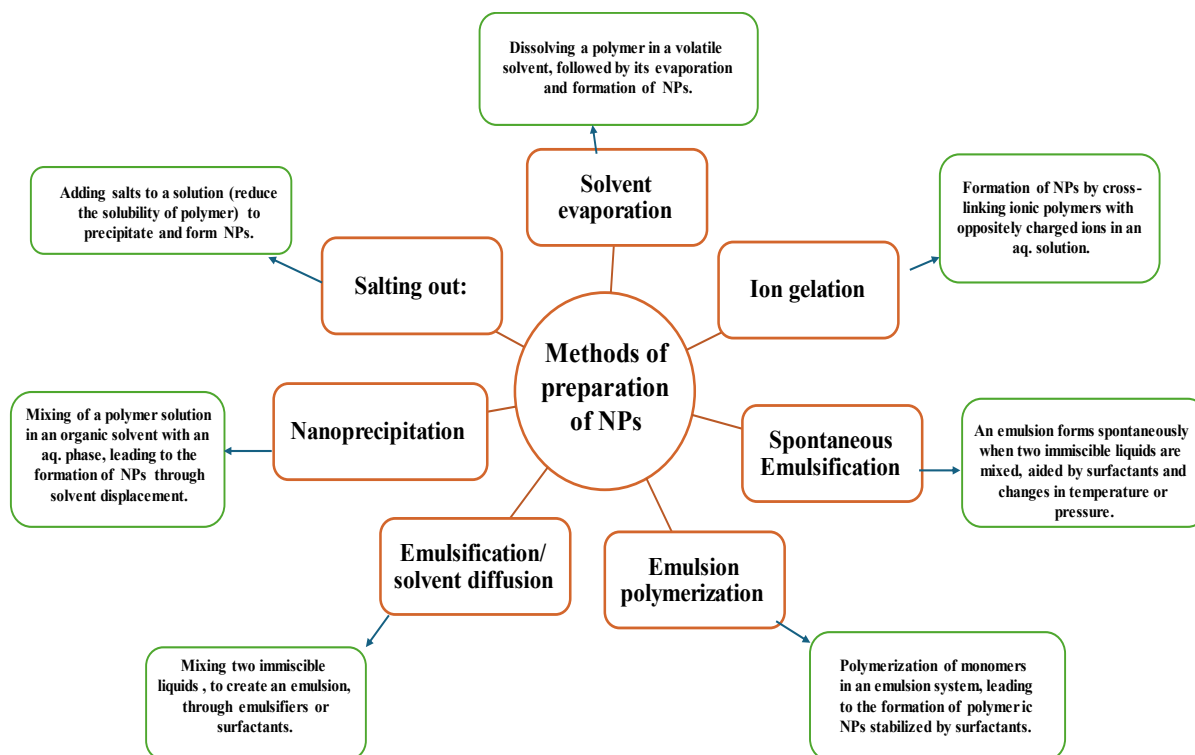


Figure 2. Methods of preparation of NPs.

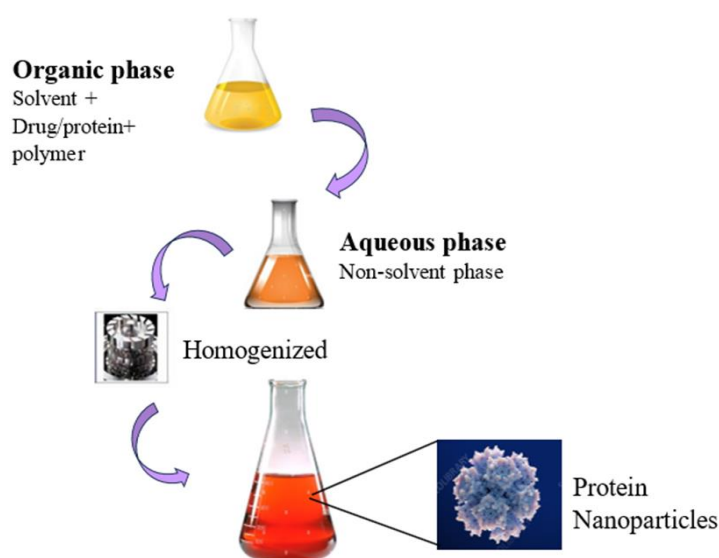


Figure 3. Preparation of NPs.

5.1. Solvent evaporation method

An emulsion is prepared by dissolving the polymer in an organic solvent like chloroform, acetone, or ethyl acetate using polyvinyl alcohol as a stabilizer. If the homogenization procedure is carried out for a long enough duration, it can help to evaporate the organic solvent [37]. Ultracentrifugation is used to gather the NPs at the end of the homogenization process. The desired particle size and other characteristics can be attained by modifying process variables, including the ratio of polymer to

organic solvent, the type of organic solvent, and the speed and duration of homogenization [38].

5.2. Spontaneous emulsification/solvent diffusion method

This method utilizes water-immiscible solvents such as dichloromethane or chloroform as the organic phase, while water-miscible solvents like methanol or acetone serve as the organic phase. NPs are formed when these two phases are combined, resulting in interfacial turbulence and leading to the formation of NPs. Despite producing nanosized particles, this approach has certain drawbacks, such as the existence of leftover organic solvent [39,40].

5.3. Salting out/emulsion diffusion method

In the salting-out process, the polymer is first dissolved in a water-miscible organic phase, such as acetone or tetrahydrofuran, and then added to the aqueous phase containing the emulsifier [39]. The fast addition of water to the emulsion, combined with gentle stirring, decreases the ionic strength, causing the water-soluble organic solvent to migrate to the aqueous phase and resulting in the formation of polymeric nanoparticles [41].

5.4. Phase separation method

This technique works with both lipophilic and hydrophilic medications. The hydrophilic medications are typically introduced to the organic phase after being dissolved in water. Conversely, the medications that are lipophilic are dissolved in polymer solutions. After the aqueous and organic phases are combined to form an emulsion, a second organic nonsolvent, such as silicone oil, is added while stirring vigorously. Silicone oil is miscible with the initial organic phase but does not dissolve the medication. Consequently, the first organic solvent is extracted, reducing the solubility of the polymer and causing phase separation, leading to the formation of a polymer coacervate. Drug-loaded nanoparticles are formed when this polymer coacervate adsorbs onto active pharmaceutical ingredients [42].

5.5. Emulsion polymerization method

Emulsion polymerization is a widely used technique for synthesizing polymeric NPs. In this process, the monomer is emulsified in a continuous phase of an immiscible liquid. A polymerization reaction occurs in situ, resulting in nanospheres. The choice between water-in-oil (w/o) or oil-in-water (o/w) emulsion systems depends on the hydrophobicity of the desired monomer. Surfactants play a crucial role in stabilizing emulsions and controlling particle size. High-speed mixing, homogenization, or ultrasound sonication are common methods for carrying out this process [43].

5.6. Nanoprecipitation

The nanoprecipitation technique utilizes two miscible solvents: One acts as a good solvent (usually an organic solvent such as acetone, isopropanol, or ethanol), while the other functions as a non-solvent for the polymer or lipid used to form the NPs. For instance, water can be a non-solvent. The procedure involves preparing an

organic phase and a non-solvent phase (often referred to as the aqueous phase). Both phases ensure complete solubility of all starting materials. The organic phase may include polymers, solid or liquid lipids, surfactants with low HLB values, and active compounds dissolved in organic solvents. The solubility of the active molecule in the solvent affects the drug loading capacity of the particles. Meanwhile, the non-solvent phase includes stabilizing agents dissolved in water, facilitating NP formation and ensuring system stability [44,45].

5.7. Ion gelation

Ionotropic gelation is a well-studied method for preparing nanocarrier systems due to its mild conditions and straightforward procedures. In this technique, polyelectrolytes (such as chitosan, alginate, hyaluronic acid, and carrageenan) are cross-linked in the presence of counterions. The cross-linking process involves the formation of a network through ionic bridges between macromolecular chains. Typically, a charged ionic entity with a defined molecular weight serves as the cross-linking agent. Researchers frequently use this procedure to prepare NPs [46,47].

6. Characterization of protein loaded NPs

6.1. Size, polydispersity index (PDI), and zeta potential

Particle size characterization of protein-loaded NPs involves morphological examination by transmission electron microscopy (TEM), scanning electron microscopy (SEM), and atomic force microscopy (AFM) [46]. Transmission electron microscopy (TEM) provides direct visualization of NP size and provides detailed images of NP morphology. Dynamic light scattering (DLS) is commonly used to measure the hydrodynamic diameter and PDI of NPs [48–50].

6.2. Stability and surface properties

Surface charge (zeta potential), hydrophilicity, and wettability are vital for determining the interactions between NPs and biological systems. Zeta potential measurements predict the stability of NP dispersions and can be measured by a zeta sizer. *This* measures the surface charge of NPs, which influences their stability in suspension. A higher absolute zeta potential value typically indicates better stability due to electrostatic repulsion between particles. NPs with zeta potential of more than 30 mV (+/–) have been reported to be stable in the deposit because the surface charge prevents the particles from clotting.

6.3. Encapsulation efficiency

The encapsulation efficiency is determined by measuring the amount of protein in the supernatant after centrifugation of the NPs. The protein content is quantified using methods like Lowry's assay or BCA assay. The encapsulation efficiency is calculated as:

$$\text{(Total protein – Protein in supernatant)/Total protein} \times 100\%$$

Release Kinetics: The drug release behaviour of NPs is evaluated in simulated biological fluids to understand how they release their therapeutic load over time. This assessment is conducted using techniques like dialysis or sample-and-separate methods.

Biodegradability and degradation rate: Gel Permeation Chromatography (GPC) and Mass Spectrometry (MS) are used to track the degradation of polymeric NPs over time.

Cellular Uptake Studies: NPs internalization by cells is measured and visualized through techniques such as confocal microscopy and flow cytometry, which help assess how effectively the NPs are delivered to target cells.

Cytotoxicity: The compatibility of NPs with cells is tested using various assays like MTT, XTT, or LIVE/DEAD assays to evaluate their potential toxicity to healthy cells.

Haemolysis and Protein Binding: Haemolysis tests determine how NPs interact with red blood cells and are analysed through SDS-PAGE or mass spectrometry that investigates how serum proteins attach to NPs and potentially affect their behaviour.

In-vivo characterization: It focuses on how polymeric NPs perform inside living organisms, providing critical information for preclinical evaluation.

- **Pharmacokinetics (PK):** The absorption, distribution, metabolism, and excretion (ADME) of NPs are examined using techniques such as HPLC, LC-MS, and fluorescence imaging. Prolonged circulation times of NPs are often preferred for better bioavailability.
- **Biodistribution:** NPs distribution across different organs or tissues is analysed using radioactive labelling or fluorescent dyes in combination with imaging technologies like MRI, PET, or bioluminescence imaging to ensure that NPs reach their target, such as tumours.
- **In-vivo toxicity:** The safety of NPs is evaluated by examining major organs (e.g., liver, kidneys, lungs) using histopathological analysis, blood tests, and immune response monitoring. Both acute and chronic toxicity assessments are conducted to detect any adverse effects.
- **In-vivo Efficacy:** These studies evaluate whether the NPs effectively deliver their therapeutic payload in animal models. Endpoints like tumour shrinkage, inflammation reduction, or changes in specific biomarkers are monitored to assess therapeutic success.
- **Immunogenicity and Immune Response:** NPs should not trigger harmful immune reactions. The immune response is gauged by measuring levels of cytokines, antibodies, and other immune markers using ELISA or multiplex assays.
- **Targeting Efficiency:** The effectiveness of targeted delivery mechanisms (e.g., ligand-receptor binding or enhanced permeability and retention (EPR) effect) is studied. Imaging techniques or tissue analysis help determine the extent of NPs accumulation at the target site [51–57].

Each of these techniques provides insights into different aspects of protein-loaded NPs, helping to optimize their design for specific applications, such as drug delivery or therapeutic interventions (**Table 5**).

Table 5. Applications.

Polymer used	Type of polymeric NP /method of preparation	Route of administration	Drug/Active agent	Treatment	Reference
Chitosan	Ion crosslinking method	Nasal	Quercetin	Allergic rhinitis (AR)	[58]
Chitosan	CS-Au based on gold NPs and chitosan (CS)	Intravenous	Myricaria germania	Immunization	[59]
Poly (β -amino esters) (PBAEs)	Encapsulation of the synthetic mRNA encoding bevacizumab, an anti-VEGF antibody in NPs	Intravenous	mRNA encoding bevacizumab	Non-small cell lung cancers (NSCLCs)	[60]
Chitosan	MTX was entrapped in the Chitosan NPs	Topical application	Methotrexate (MTX)	Rheumatoid arthritis	[61]
Chitosan	Encapsulation of AMP NRC-07 in CS-NPs by ionotropic gelation	Intravenous	antimicrobial peptide NRC-07	Antibacterial and <i>in vitro</i> anticancer activities	[62]
Elastin-like peptides (ELPs)	Supramolecular NPs based on elastin-like peptides modified capsid protein	Intravenous	Doxorubicin	Murine melanoma and colorectal cancer.	[63]
Poly(N-isopropylacrylamide)	Temperature-responsive Polymer NPs	Intravenous	Paclitaxel	Anticancer activity	[64]
Albumin	Elastin-targeted NPs	Intravenous	Doxycycline	LPS-mediated lung inflammation	[65]
Polymeric and lipid-based	Levodopa-loaded NPs	Intravenous, transdermal delivery and intranasal administration	Levodopa	Parkinson's disease	[66]
Hydrogel	pH-responsive polymeric nanocarriers	Intravenous	IL-12	Immunotherapy of Cancer	[67]
Chitosan	Concanavalin A (ConA) coated chitosan (CS) nanocarrier	Intravenous	Short antimicrobial peptide (CM11)	Helicobacter pylori gastric infection	[68]
Glutenin	Glucose-conjugated glutenin NPs	Intravenous	Camptothecin	Breast cancer	[69]
Chitosan	Peptides in chitosan NPs Coated with Zein	Oral	Antihypertensive Peptides, Isoleucine-Proline-Proline and Leucine-Lysine-Proline	Antihypertensive activity	[70]
ROS-responsive polymer	Modified emulsion approach	Intravenous	Dexamethasone	Acute lung injury (ALI)	[71]
Chitosan	Hyaluronic acid coated chitosan NPs	Oral	Insulin	Hypoglycemic activity	[72]
Phenylboronic ester	Phenylboronic ester-modified polymeric NPs	Nano vaccine	TRP2 peptide antigen delivery	Cancer immunotherapy	[73]
Poly(lactic-co-glycolic) acid (PLGA)	Double emulsion technique	Oral	Capreomycin peptide	Impact of stress conditions on peptide degradation: Thermal, mechanical, chemical	[74]
Chitosan	Self-assembled chitosan NPs by ionic cross-linking technique	Intranasal	Recombinant protein interleukin-17 receptor C (IL-17RC)	Asthma	[75]
pH-sensitive polymer	D-melittin polymeric NPs	Intravenous	D-melittin	Anti-cancer treatment	[76]
Chitosan	Cross-linked NPs	Percutaneous delivery	Betamethason	Contact dermatitis	[77]

Table 5. (Continued).

Polymer used	Type of polymeric NP /method of preparation	Route of administration	Drug/Active agent	Treatment	Reference
Hypoxia-responsive polymer PLA	Targeted and non- targeted self-assembled polymeric NPs	Intravenous	Doxorubicin (DOX)	Hypoxic, triple-negative breast tumors	[78]
Chitosan	Self-gelation method	Oral	Insulin	Diabetes	[79]
Poly (lactic-co-glycolic acid) (PLGA)	Double emulsion modified method	Intravenous	Recombinant adrenomedullin-2	Angiogenesis	[80]
PLGA, PEG	S2P peptide-conjugated PLGA-Maleimide-PEG NPs, modified emulsion/solvent evaporation technique.	Intravenous	Imatinib	Atherosclerosis	[81]
Poly (lactide-co-glycolide)-b-poly (ethylene glycol) NPs	Polymeric NPs functionalized with muscle-homing peptides	Intravenous	Phosphatase and tension homology inhibitor to skeletal muscle	Duchenne muscular dystrophy (DMD)	[82]
Human serum albumin (HSA)	Synthetic protein NPs (SPNP), polymerized HSA equipped with iRGD	Intravenous	siRNA against Signal Transducer	Glioblastoma	[83]

7. Conclusions and future prospects

Peptides and proteins are essential for numerous biological reactions and play significant roles in various pathological conditions. However, their therapeutic application in treating life-threatening disorders encounters several challenges, including instability, poor absorption, enzymatic degradation, a short biological half-life, and rapid elimination. Polymeric NPs have shown considerable potential in enhancing the absorption of macromolecules. These polymers are typically inert, biocompatible with biological fluids, biodegradable, and can be removed from the body as inert biodegradable products. The choice of polymers used to deliver therapeutic proteins and peptides significantly impacts their therapeutic efficacy. Polymeric NPs hold promise for various delivery routes, including nasal, pulmonary, oral, and ocular delivery; nonviral gene delivery; and crossing the blood-brain barrier. A versatile system that can ensure the delivery and systemic stability of various proteins and peptides would be highly beneficial in the near future.

Author contributions: Conceptualization, NM and NS (Nawale Sneha); methodology, NM; software, NS (Neerude Sirisha); validation, NM, NS (Nawale Sneha) and NS (Neerude Sirisha); formal analysis, NM; investigation, NS (Nawale Sneha); resources, NS (Neerude Sirisha); data curation, NM and NS (Nawale Sneha); writing—original draft preparation, NS (Neerude Sirisha); writing—review and editing, NM and NS (Nawale Sneha); visualization, NS (Neerude Sirisha); supervision, NM; project administration, NM; funding acquisition, NS (Neerude Sirisha). All authors have read and agreed to the published version of the manuscript.

Conflict of interest: The authors declare no conflict of interest.

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