

# A Comprehensive Review on Nanoplex Nanotechnology Strategies

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One strategy used to get around the problems with conventional medication delivery systems is the use of nanotechnology based on the creation and growth of nanostructures. The newest development in nanotechnology is the creation of Nanoplex. A polyelectrolyte with an oppositely charged drug nanoparticle forms a complex known as a nanoplex. Both cationic and anionic medicines combine with polyelectrolytes that have opposing charges to produce complexes. The yield of Nanoplex is higher and the complexation efficiency is better when compared to other nanostructures. Additionally, nanoplex are simpler to make. Using scanning electron microscopy, differential scanning calorimetry, X-ray diffraction, and dialysis investigations, the production yield, complexation efficiency, drug loading, particle size, and zeta potential of nanoplex formulation are all evaluated. Nanoplex have a wide range of uses in various domains, including cancer treatment, medication delivery to the brain, drug delivery via genes, and drug delivery via proteins and peptides.

**Keywords:** Application; Evaluation; Nanotechnology; Nanoplex; Mechanism.

The Latin-based origin of the word “Nano” is “dwarf” (Rangasamy et al., 2011). The field of study known as nanotechnology is concerned with molecular processes and the nano length scale (Kasar et al., 2018). Since several decades, the term “nanotechnology” has been most frequently used in the sciences of electronics, physics, engineering, pharmaceutical and biomedical disciplines, however, have not yet been thoroughly studied (Rangasamy et al., 2011). It has made a significant contribution to a number of medical specialties, including gene delivery, brain targeting, and the development of oral vaccines, as well as to the domains of cardiology, immunology, ophthalmology, oncology, endocrinology, and

pneumology (Kadam et al., 2015). For better pharmaceutical applications, nanotechnology offers intelligent materials, systems, and devices (Bhattacharyya et al., 2009). This review will emphasize on a nanoplex.

## **Nanoplex: An Innovative Development in Nanotechnology**

A drug nanoparticle compound with an electrostatically charged polyelectrolyte is known as a nanoplex. To form a nanoplex, an oppositely charged polyelectrolyte and a cationic or anionic medication must react. The majority of potential medication candidates have low-saturation solubility in the aqueous phase. Because many medications have a low oral bioavailability,

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research is currently focused on making them more soluble. As a result, frequent dosing is necessary, which places a financial and pharmaceutical load on patients. Three main strategies can be used to increase bioavailability: (1) converting the API into a salt form that is extremely soluble, (2) creating crystalline nanoparticles (or nano API) of the API, and (3) delivering the API in its amorphous form (Ando, Radebaugh 2005). The simplest method for boosting a medicine's saturation solubility is to create a salt of the drug using a mild organic acidic or basic. However, salt production does not ensure an increase in saturation solubility (Rabinow *et al.*, 2004). Due to the fact that the particles are reduced to nano size and behave in accordance with the Ostwald-Freundlich solubility principle, nano API formulations aren't restricted to for acidic or basic medications.

According to the Ostwald-Freundlich equation, the effect of nanoionization on saturation solubility is only felt at diameters much larger than 100 nm (Grant *et al.*, 1995). It has been discovered that nanoscale API with diameters between 150 and 200 nm only have a 15% higher saturation solubility than their microscale counterparts. For the production of nano API with a size less than 100 nm, the current nano API formulation processes (such as high-pressure homogenization, wet milling) are unpromising. Consequently, improvements in nano API preparation methods are required to increase bioavailability. Making the API's amorphous form metastable is another tactic for boosting apparent solubility. The amorphous API dissolves into a highly supersaturated solution with an apparent solubility that is considerably higher than the saturation solubility of the crystalline equivalents. If the high supersaturation level can be maintained for a long enough time to allow for absorption, it will promote medication absorption throughout the gastrointestinal lumen and increase bioavailability. The important aspect of amorphous forms is that it has been demonstrated that high supersaturation levels result in increased bioavailability *in vivo* (Yang, Johnston, Williams 2010, Tam *et al.*, 2008).

#### **Mechanism of nanoplex formation**

To create an anionic or cationic drug solute, the water-insoluble medication must first dissolve in an acidic or basic solution. The drug-polyelectrolyte electrostatic interaction and charge

neutralization are then triggered by the subsequent mixing of the ionized drug solution with a polyelectrolyte solution that is negatively charged. The drug solute undergoes a transformation that returns it to its minimally soluble state upon charge neutralization, which causes a loss of solubility. As a result, there is fast precipitation and the creation of the drug-polyelectrolyte complex at the nanoscale. The inability of the drug molecules to assemble into organized crystalline structures is due to a combination of fast precipitation and powerful electrostatic interactions between the drug and the polyelectrolyte. The result is the formation of an amorphous drug-polyelectrolyte nanoparticle combination (Cheow *et al.*, 2012).

Methods which use in to making nanodrug from hydrophobic drug are as follows.

#### **Emulsified solvent evaporating technique**

In this nanoparticle formulation approach, a hydrophobic drug and a polymer are dissolved in an immiscible solvent. This solvent mixture is then carefully introduced into an aqueous phase containing a dissolved surfactant in a dropwise manner, followed by the application of homogenization. After solvent removal, achieved through lyophilization, nanoparticles are obtained. This method involves the addition of a substantial amount of aqueous phase into the solvent, promoting the diffusion of the organic solvent from the internal phase to the external phase. For certain drugs, a double emulsion method is favored. Successful implementation of this technique requires meticulous observation of homogenization duration and intensity, as well as a thoughtful consideration of the specific drug and polymer employed. This versatile method is applicable not only to hydrophobic drugs but also to water-soluble ones, utilizing a double emulsification method where the drug is initially solubilized in an aqueous solution. The choice of polymer, solvent type, and homogenization intensity plays a crucial role in the effectiveness of this formulation strategy. (Sivasankar *et al.*, 2010)

#### **Direct compression method**

Nanoparticle powder, derived from a nanosuspension through techniques like spray drying, offers a convenient oral administration method by filling capsules. For acid-sensitive drugs, the nanoparticle powder finds placement in hard gelatin capsules. An alternative oral

**Table 1.** Types of Nanostructures

Type of Nanostructure	Description	References
Nanosuspension	Pharmaceutical nanosuspensions are stabilised, heterogeneous aqueous dispersions containing drug particles that are insoluble at the nanoscale due to surfactants.	Jacob et al., 2020
Solid lipid nanoparticles	Solid lipid nanoparticles (SLN) are aqueous colloidal dispersions with solid biodegradable lipids as their matrix. SLN formulations have been developed and thoroughly characterised in-vitro and in-vivo for a variety of administration routes, including parenteral, oral, cutaneous, ocular, pulmonary, and rectal	Lingayat et al., 2017; Garud et al., 2012
Nanofibers	Fibres with a diameter between 50 and 500 nanometres are referred to as nanofibers. According to the National Science Foundation (NSF), nanofibers are materials with at least one dimension of 100 nanometres (nm) or less. In recent years, nanofibers have become a popular drug delivery technology in the healthcare industry for a number of disorders.	Kattamuri et al., 2012
Nanocomposites	Nanocomposites are multi-phase materials with at least one phase with diameters between 10 and 100 nm. Among other nanocomposites, polymer-based nanocomposite (PNCs) has emerged as a significant field of current study and development.	Pandey et al., 2017
Carbon nanotubes	Carbon nanotubes (CNTs) are beneficial in the disciplines of nanotechnology and medicines due to a variety of new features they possess. They exhibit a wide spectrum of electrical, thermal, and structural properties and are nanometres in diameter and several millimetres in length. CNTs are allotropes of carbon. They are tubular in shape, made of graphite.	Hirlekar et al., 2009
Nanopores	Due to their exceptional qualities in terms of thermal insulation, controllable material separation and release, and suitability as templates or fillers for chemistry and catalysis, materials with pore sizes in the nanometre range are of particular interest for a wide range of industrial applications.	Wanunu et al., 2012
Liposomes	In clinical settings, liposomal drugs have been shown to be most helpful when their average diameter is in the range of ultra filterable (b200 nm in diameter), when they are able to “passively” accumulate at sites of increased vasculature permeability, and when they are able to lessen the side effects of the encapsulated drugs compared to free drugs.	Allen et al., 2013
Polymeric nanoparticle	Solid colloidal particles having a diameter ranging from 1 nm to 1000 nm are called polymeric nanoparticles. Due to their distinct characteristics and behaviours brought on by their small size, polymeric nanoparticles (PNPs) have garnered a lot of attention in recent years.	Jawahar et al., 2012; Crucho et al., 2017
Polymeric micelles	Nanoscale drug delivery devices known as polymeric micelles have a core-shell structure that forms when amphiphilic block copolymers self-assemble in aqueous solution. Polymeric micelles, or aggregation colloids created in solution by the self-assembly of amphiphilic polymers, are an inventive way to solve a number of problems with medication administration, such as their low water solubility and poor permeability	Ghezzi et al., 2021

	through biological barriers.	
Dendrimers	Dendrimers are distinctly shaped nanoparticles of dimensions about 1–15 nanometres. It has extremely flexible surface functionalization and are highly regarded pharmaceutical. They are branch-like macromolecules with a central core unit that exhibits excellent molecular homogeneity, a limited molecular weight dispersion, distinctive size and shape properties, and a highly functionalized terminal surface.	Mishra et al., 2011
Nanocapsules	The term “polymeric nanoparticles” refers to delivery systems with dimensions less than 1 µm, which are also known as “nanocapsules” or “nanospheres” according to their make-up. Nanospheres have a matrix organisation of polymer chains, and the drug molecules can be distributed, entrapped or dissolved in, adsorbed, or chemically bonded to the polymer matrix, whereas nanocapsules often have an oil core that leads to a vesicular or membrane-type structure.	Erdogar et al., 2018
Nanoplex	A drug nanoparticle compound with an electrostatically charged polyelectrolyte is known as a nanoplex. To create a nanoplex, a medication that is either cationic or anionic is made to react with a polyelectrolyte that has an opposing charge.	Kadam et al., 2015
Magnetic nanoparticles	A type of manufactured particulate materials with a particle size of less than 100 nm that can be controlled by external magnetic fields is known as magnetic nanoparticles. Dextran, a polymer, serves as the core of magnetic nanoparticles, which are then coated in an inorganic material like iron oxide.	Indira et al., 2010
Nanoemulsions	Thermodynamically stable, transparent (translucent) dispersions of oil and water with interfacial layers of surfactant and cosurfactant molecules are known as nanoemulsions or submicron emulsions. Their droplets are smaller than 100 nm in size.	Indira et al., 2010
Quantum dots	Quantum dots (QDs) are described as particles with physical dimensions smaller than the exciton Bohr radius and are nanometre-scale semiconductor crystals made of elements from groups II-VI or III-V.	Jamieson et al., 2007
Carbon allotrope graphene's	Pure carbon is used to make graphene, which has atoms arranged in a sheet that is one atom thick and has a regular hexagonal structure resembling that of graphite. It is a type of carbon known as an allotrope, and its crystal structure is made up of a single planar sheet of tightly packed, sp <sup>2</sup> -bonded carbon atoms. Consequently, graphene's can be thought of as the precursor to graphite, fullerene, and CNTs.	Bera et al., 2010

delivery strategy involves the transformation of the nanoparticle powder into tablet form. Here, the drug nanosuspension is blended with a matrix-forming substance, such as micro-sized polymer powder or lipid, combined with lactose powder. Following spray drying, the liquid phase converts into an API-matrix compound, resulting in a free-flowing powder. This powder undergoes direct compression, yielding extended-release tablet formulations. (Sivasankar et al., 2010)

#### Advantages of nanoplex

1. The process for making Nanoplex is easy because it simply requires the mixing of two solutions, one each of the medication and polyelectrolyte.
2. It is not necessary to use a lot of solvents during the preparation of nanoplex.
3. In comparison to other nano formulations, the synthesis of nanoplex requires very less energy.
4. It takes a short period for nanoplexes to form.
5. For the preparation of Nanoplex, complex equipment is not required.

### **Evaluation of nanoplex Complexation efficiency**

This is described as the mass of a drug that, in relation to the drug that was first added, forms a complex with the polyelectrolyte. The optical density of the supernatant following the initial centrifugation of the nanoplex suspension is used to compute it

- **Production yield:** It is the proportion of the dry nanoplex generated after freeze-drying to the weight of the medication and polyelectrolyte initially used
- **Drug loading:** This represents the real drug content of the nanoplex powder. It is computed by combining 5 mg of the nanoplex powder with 20 ml of ethanol, centrifuging the mixture, and measuring the absorbance of the resulting solution
- **Particle size analysis:** A particle size analyzer is used to determine the nanoplex's particle size.
- **Zeta potential:** A zetasizer is used to determine the nanoplex's zeta potential.
- **Differential scanning calorimetry (DSC):** To ascertain how a medicine interacts with a polyelectrolyte, DSC is used. Their melting temperatures are also used to determine the interactions, and a DSC instrument is used to calculate the difference. Graphs are used to display the results.
- **Powder X-ray diffraction:** A powder X-ray diffractometer is used to determine the patterns of samples' powder X-ray diffraction. This provides information regarding the sample's nature.
- **Scanning electron microscopy:** Using a scanning electron microscope, the sample's surface morphology is examined.
- **Dissolution study:** The dialysis bag method is used to assess the dissolution of the nanoplex to ascertain the drug release.
- **Saturation solubility study:** The orbital flask shaker method is used to assess both the drug's and the formulation's solubility. Through the use of spectrophotometric analysis, the drug concentration is ascertained from the absorbance.
- **Stability study:** By putting the nanoplex in an environmental stability chamber, it is tested for stability, and its drug content is measured.

### **Applications of Nanoplex**

#### **Enhancement of the solubility and dissolution rates of medicines**

Class II and IV medicines, which are

poorly water-soluble, can be prepared as Nanoplex to improve their bioavailability by increasing their solubility and rate of dissolution. An increase in solubility lowers a drug's dosage required (Cheow *et al.*, 2012).

#### **Drug delivery to the brain**

The most significant hurdle to the creation of a new medicine for the central nervous system is the blood-brain barrier (BBB). Endothelial cells with tight connections, enzymatic activity, and active efflux transport mechanisms, which are relatively impermeable, characterize the BBB. Through the action of enzymes or efflux pumps, it successfully blocks the entry of water-soluble molecules from the blood circulation into the CNS and can lower the concentration of lipid-soluble molecules in the brain. As a result, the BBB only allows the selective transit of chemicals required for proper brain function. A number of disorders, including HIV-1, AIDS, dementia, and cerebral ischemia, have been linked to nanoparticles, which have been proposed as non-viral gene delivery vectors and as having tremendous promise for therapeutic usage. The effectiveness and specificity of MMP-9-siRNA quantum dot (QD) complexes (nanoplex) in reducing the expression of the MMP-9 gene in the brain microvascular endothelial cells (BMVECs) that make up the BBB have been assessed. Adela Bonoiu *et al.* showed how to modulate MMP-9 activity in BMVECs and other MMP-9-producing cells using a unique nanoplex siRNA delivery technique. This application will stop neuroinflammation and protect the BBB (Bonoiu *et al.*, 2009, Fang *et al.*, 2023).

#### **Nanoplex for gene delivery**

Due to its biocompatibility, biodegradability, low cytotoxicity, absence of pathogenicity, and low immunogenicity, non-viral vector-mediated gene therapy is now one of the most alluring approaches used. Currently, research is concentrated on creating non-viral vectors made of cationic polymers, liposomes, and dendrimers. DNA's anionic character has been found to be usefully neutralized by cationic polymers, which effectively condenses DNA and makes it easier for DNA to enter cells. Plasmid DNA delivery formulations use an intriguing family of vectors called cationic polymers (Thomas *et al.*, 2010, Wada *et al.*, 2023).

### Drug targeting in cancer treatment

For non-viral vectors, a wide variety of cationic lipids have been synthesized. A cationic lipid typically consists of three parts: a hydrophobic lipid anchor group that aids in the formation of the micellar structure and can interact with cell membranes; a linker group, such as an ester, an amido group, or a carbamate; and a positively charged head-group that is primarily made up of cationic amines. As the anchor group, cationic cholesterol derivatives might be chosen due to their strong transfection activity and low toxicity. In cationic cholesterol derivatives, the linker group regulates the conformational flexibility, level of stability, biodegradability, and effectiveness of gene transfection. When the nanoparticle/siRNA complex (nanoplex) is created in a NaCl solution, cationic nanoparticles made of OH Chol (NP-OH) could deliver siRNA with a high transfection efficiency in vitro (Hattori *et al.*, 2008, Akkin *et al.*, 2023).

### Drug delivery of proteins and peptides

Numerous bioactive compounds and vaccines based on peptides and proteins have been discovered as a result of significant advancements in biotechnology and biochemistry. The development of appropriate carriers is still difficult since the gastrointestinal tract's epithelial barrier restricts the bioavailability of these compounds and makes them vulnerable to gastrointestinal breakdown by digestive enzymes. Proteins are a good choice for nanoplex formulation because they are charged molecules that form a complex with polyelectrolytes (Woitiski *et al.*, 2007; Ranjan *et al.*, 2010).

### CONCLUSION

The majority of medications taken orally have an amphiphilic character and are soluble in a mild acid or base. The straightforward complexation procedure can convert them into amorphous Nanoplex. A nanoplex can be created by simply combining two solutions under room temperature. It is also quick and free of solvents. It creates uniform-sized nanoparticles with good complexation efficiency, drug loading, and production yield while using little energy.

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### Conflict of Interest

The authors declare no conflict of interest, Financial or otherwise

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