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NANOSTRUCTURED LIPID CARRIERS: A REVOLUTIONARY PATH FOR TARGETED DRUG DELIVERY AND DISEASE MANAGEMENT

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ABSTRACT

Nanostructured lipid carriers (NLCs) are gaining significant attention in both cosmetics and pharmaceuticals due to their versatile properties. In cosmetics, NLCs are utilized to enhance the delivery of active ingredients, such as vitamins and antioxidants, ensuring deeper skin penetration and improved effectiveness. They also offer controlled release, protecting sensitive compounds from environmental degradation caused by UV exposure and oxidation. In pharmaceuticals, NLCs provide a promising solution for the delivery of poorly soluble drugs. They improve bioavailability, protect sensitive molecules, and allow for targeted delivery to specific sites, minimizing side effects. NLCs are particularly useful for delivering a range of therapeutic agents, including anticancer drugs and antibiotics, enabling more efficient treatment options. A review related to NLC technology highlights ongoing innovations in drug delivery systems and patents focus on developing novel formulations, enhancing drug encapsulation techniques, and optimizing release profiles. This reflects the growing interest in NLCs as a means to improve the performance and stability of both pharmaceutical and cosmetic products. Despite their potential, several challenges remain. Issues like large-scale production difficulties, formulation consistency, regulatory approval hurdles, and concerns about skin irritation or toxicity need to be addressed. Furthermore, the long-term stability of drugs encapsulated within NLCs continues to be a challenge. Looking ahead, future developments in NLC technology are focused on improving targeting mechanisms, enhancing stability, and integrating NLCs with advanced technologies such as nanogels. These advancements could further optimize drug delivery and expand NLC applications in personalized medicine and advanced cosmetic treatments.

KEYWORDS: Nanotechnology, Nanostructured lipid carriers (NLCs), Targeted drug delivery, Lipid nanoparticles, pharmaceutical delivery.

INTRODUCTION

In recent years, it has become clear that developing novel medications alone is insufficient to assure significant advances in pharmacological therapy. Experiments with promising results in vitro are frequently followed by unsatisfactory results when tried in vivo or therapeutically. The most common reasons for these failures are diverse. One key cause is a lack of drug concentration in the body due to quick metabolism, which reduces the medicine's effectiveness. Furthermore, widespread distribution leads to increased drug toxicity, which has negative consequences. Poor formulation solubility and wide variations in plasma drug levels between people are also important impediments to establishing consistent treatment outcomes. To address these issues, the development of effective medication delivery devices has received increased attention. Researchers have spent decades developing nanosized drug-carrier systems to increase medication stability, bioavailability, and targeting. These drug delivery technologies are roughly classified into two types: polymeric nanoparticles and lipid nanoparticles. Polymeric nanoparticles are solid colloidal particles formed from either non-biodegradable synthetic polymers or biodegradable macromolecules obtained from synthetic, semi-synthetic, or natural origin. Despite their advantages, polymeric nanoparticles have some substantial downsides, including polymer cytotoxicity and a lack of scalable production processes for large numbers. Nanotechnology is used in many fields, including environmental science, medicine, cosmetics, and nutraceutical research. Nanotechnology has become a powerful tool in recent years for tackling the limitations of traditional drug delivery methods. Nanocarriers are colloidal systems with an average diameter of less than 1 micron. Since they have special qualities, such as a high surface-to-volume ratio and nanoscale size, nanoparticles can be employed for medicinal purposes. Their decreased size compared to biological macromolecular medicines or standard chemotherapeutic agents allows them to be coupled with several support components and pharmaceutically active chemicals. These components enable stimulus-based activation, imaging, targeting, and degradation resistance. However, the body processes nanoparticles differently than it do conventional medications. Nanoparticles have distinct biodistribution profiles and hydrodynamic characteristics. It is noteworthy that interactions occurring at the nano biological level have the potential to be used for better drug delivery. The encapsulating moieties of nanocarriers can be altered to improve their pharmacokinetic and biodistribution characteristics, decrease toxicity, regulate

release, improve solubility and stability, and deliver their payload to targeted sites. The physiochemical characteristics of nanocarriers, such as their surface, composition, and shape, can be altered to improve their activities with fewer side effects. Nanostructured lipid carriers (NLCs) present promising advancements in drug delivery systems, yet they face several challenges that hinder their clinical application. These challenges include formulation complexities, stability issues, and biological barriers. Examples of nanocarriers include polymeric, lipidic, inorganic nanoparticles, liposomes, nanotubes, nanocomplexes, noisome, and other forms. The surface properties of nanocarriers play a crucial role in determining their bioavailability, stability, cellular uptake, and biodistribution. The zeta potential, which reflects the surface charge, affects the tendency of nanocarriers to aggregate, suggests possible electrostatic interactions among them, and aids in selecting appropriate coating materials. The shape and aggregation behavior of nanocarriers also influence several biological factors, such as their half-life, targeting efficiency, and toxicity. Various non-spherical shapes, including cubes, cones, hemispheres, cylinders, and other complex forms, significantly affect these biological functions. Lipid nanoparticles, which are manufactured from natural or biocompatible materials, are a possible option. They pose a lower toxicity risk than polymeric nanoparticles, owing to their natural and biological origins. This makes lipid nanoparticles a safer option for drug delivery, though challenges still exist in optimizing their design and production. Lipid nanoparticles with a solid matrix, known as Solid Lipid Nanoparticles (SLNs), are derived from oil-in-water nano emulsions, where liquid oils are replaced with solid lipids. SLNs were first developed in the early 1990s. One of their key advantages is the use of physiological lipids, which avoids the need for organic solvents and allows for large-scale production. As drug delivery systems, SLNs can improve bioavailability, protect sensitive drugs from harsh environments, and offer controlled drug-release properties. However, SLNs also present certain limitations, including unpredictable gelation tendencies, polymorphic transitions, and low drug incorporation due to the crystalline nature of the solid lipids. To address some of these challenges, **Nanostructured Lipid Carriers (NLCs) were developed around the turn of the millennium. NLCs are created by blending solid lipids with liquid oils, which results in a more complex matrix structure. This modification allows NLCs to overcome some of the drawbacks associated with SLNs, such as limited drug-loading capacity and drug expulsion during storage. The new generation of lipid nanoparticles, NLCs, offers a more efficient and stable drug delivery solution. This review aims to highlight the advancements in NLCs for drug delivery, particularly in targeting applications. It also explores the various types of NLCs used for drug delivery and

the different routes through which these systems can deliver therapeutic agents. Lipid nanoparticles are composed of triglycerides, partial glycerides, fatty acids, and waxes, which are blended with various surfactant mixtures to form the particle structure. These lipid nanoparticles are highly effective for targeted drug delivery because their particle size is generally less than 1 µm, which enhances their ability to deliver drugs precisely to specific sites in the body. On the other hand, polymer nanoparticles are created from biodegradable and biocompatible polymers, making them ideal candidates for the development of nanoscale drug carriers. Their biodegradability and biocompatibility allow for the controlled release of drugs over extended periods, protecting the active compounds from degradation and improving therapeutic outcomes. This characteristic makes them particularly attractive for drug delivery systems. Furthermore, the use of polymer nanoparticles can significantly improve the adsorption efficiency of drugs, as well as the surface properties of the carrier, which can lead to enhanced drug loading and better performance in terms of drug release and stability. Polymeric micelles are self-assembling carriers made from block copolymers, featuring a core-shell structure. The key characteristics of these micelles, including their size, shape, and essential micelle concentration, can be precisely controlled by adjusting the structural and physical properties of the copolymers used to create them. Inorganic nanocarriers, such as gold nanoparticles, carbon nanotubes, quantum dots, mesoporous silicon, and magnetic nanoparticles, are gaining attention for their innovative applications. These carriers are particularly useful for a range of advanced applications, including cell labeling, imaging, biosensing, targeting specific cells or tissues, and diagnostic purposes. Their unique properties, such as conductivity, magnetism, and optical characteristics, make them ideal for use in cutting-edge diagnostic and therapeutic technologies.

LIPID CARRIER-BASED DRUG DELIVERY SYSTEM

Lipid-based drug delivery systems are gaining significant attention for their ability to enhance drug therapy by providing pharmaceutical protection, controlled release, and targeted drug distribution. These systems are composed of biodegradable and biocompatible lipids, which are capable of encapsulating a wide range of therapeutic substances such as growth factors, gene therapies, cytokines, and other biologically active compounds. Unlike traditional drug delivery methods, lipid-based systems can incorporate various types of drugs, including proteins, peptides, nucleic acids, and new chemical entities, delivering them directly to specific cells or tissues to enhance therapeutic outcomes. The two main categories of lipid-based delivery systems are vesicular systems and lipid nanoparticles. Vesicular drug delivery

systems, such as liposomes, noisome, and transferosomes, are structured into highly organized layers formed by amphiphilic molecules. These molecules self-assemble in water to create concentric bilayers that encapsulate the drug, offering protection and targeted delivery. These vesicles typically range in size from 40 to 800 nm and are highly efficient in delivering drugs to deeper layers of the skin, improving skin penetration and drug absorption.

Liposomes are particularly well-known for their ability to encapsulate both hydrophilic and hydrophobic drugs within their bilayer structure, mimicking the natural plasma membrane of cells. This makes them an ideal, safe vehicle for drug delivery. Their versatility has led to their use in a variety of therapeutic applications, including anticancer treatments, gene therapy, and vaccine delivery. Noisome, made from nonionic surfactants combined with cholesterol, offer an alternative to liposomes by overcoming stability and sterilization issues while still providing similar benefits, including enhanced bioavailability and targeted delivery.

Transferosomes, developed in 1991, are a newer class of vesicles with enhanced flexibility due to the inclusion of an edge activator. This feature makes them more deformable, allowing them to penetrate biological membranes more easily. This deformability gives transferosomes a significant advantage in transdermal drug delivery over other vesicular systems.

Ethosomes are another important innovation designed to enhance transdermal drug delivery. By incorporating ethanol into the lipid bilayer, ethosomes gain increased flexibility, which helps them penetrate the skin more effectively, delivering drugs into deeper layers. This flexibility is achieved by reducing the lipid multilayer density and increasing the mobility of lipid molecules in the skin's outer layer, the stratum corneum.

In the past 20 years, the development of lipid-based nanocarriers has gained momentum, with researchers focusing on overcoming the limitations of polymeric nanoparticles, such as their cytotoxicity and manufacturing challenges. Solid lipid nanoparticles (SLNs) are one such example, designed to offer stability, biocompatibility, and protection from drug degradation. These nanoparticles are made from solid lipids and have a size range of 50 to 1000 nm, which helps to provide long-term stability and controlled release of drugs. The unique composition of SLNs and their ability to remain stable in an aqueous environment makes them ideal for applications where sustained drug release is needed.

Nanostructured lipid carriers (NLCs), which combine solid and liquid lipids in different ratios, offer enhanced stability, improved drug loading, and controlled release properties. Their smaller size and unique composition allow them to better interact with the skin, improving the drug penetration process and ensuring more efficient drug delivery to the target site. Additionally, NLCs can protect sensitive drugs from degradation due to light, oxidation, or hydrolysis, making them an ideal choice for drugs that require enhanced stability during delivery. Overall, lipid nanoparticles hold great promise for improving drug solubility, bioavailability, pharmacokinetics, and skin or ocular absorption. By improving drug delivery across physiological barriers, lipid-based systems help to reduce side effects and enhance the therapeutic efficacy of the drugs they carry, making them a valuable tool in modern medicine. Inorganic nanocarriers are a class of nanomaterials that include gold nanoparticles, magnetic nanocarriers, carbon nanotubes, quantum dots, and mesoporous silica. These carriers are preferred for drug delivery due to their malleability, large surface area, distinct crystal structures, and ease of chemical modification. Their high-density surface allows for effective binding of ligands, which is essential for targeting specific cells or tissues. Additionally, the size, shape, and composition of inorganic nanoparticles can be tailored to meet specific therapeutic requirements, such as enhancing drug loading capacity, ensuring biocompatibility, extending circulation time, and improving cellular absorption. Gold nanoparticles (AuNPs) are particularly noteworthy for their unique physicochemical and optical properties, making them ideal for both diagnostic imaging and therapeutic delivery. They can be easily functionalized with various ligands or polyethylene glycol (PEG)-containing linkers to facilitate targeted delivery of drugs or nucleic acids. A key feature of AuNPs is their ability to interact with light via surface plasmon resonance (SPR), which has drawn significant attention for its potential in cancer treatment. Gold nanoparticles are used in a variety of biomedical applications, including sensitive biomolecular screening, photothermal therapy for cancer cell destruction, and cellular drug delivery. Magnetic nanoparticles, particularly iron oxide (Fe3O4), have been approved by the U.S. FDA for use in drug delivery and clinical imaging. Their magnetic properties allow for visualization using magnetic resonance imaging (MRI), making them suitable for MRI-based imaging applications. Additionally, magnetic nanoparticles are utilized in temperature-sensitive drug release systems, as they can generate heat when exposed to an external magnetic field, enabling controlled drug release. Carbon nanotubes (CNTs), which are hollow, ordered graphitic structures, possess a range of desirable properties, including a large surface area, high strength, and low weight. These tubes, which have diameters ranging from 1 to 100 nm, are chemically stable and inert. CNTs

are particularly versatile for drug delivery due to their ability to load large amounts of therapeutic agents within their hollow cores. Furthermore, their unique structure allows for targeted, controlled release of medications, enhancing the effectiveness of therapeutic compounds. Mesoporous silica nanoparticles (MSNs) have emerged as an innovative solution for drug delivery due to their well-defined, mesoporous structure. MSNs offer a combination of chemical stability, surface activity, biocompatibility, and the ability to deliver a variety of pharmacologically active compounds. Their robust Si-O bonds make them resistant to mechanical stress and environmental degradation, ensuring their durability. One of the key advantages of MSNs is their functionalize silanol-containing surface, which enables efficient drug loading and controlled release. This unique characteristic makes MSNs particularly useful for targeted drug delivery and sustained-release applications.

Dendrimer

Dendrimers have emerged as a prominent class of nanostructured carriers, driving innovation in nanomedicine for the treatment of various diseases. These nanomaterials are synthesized from branched monomer units, with the flexibility to select different building blocks, branching units, and surface functional groups. This design allows for the customization of dendrimers in terms of size, structure, density, and solubility. Moreover, dendrimers can incorporate organic molecules and polymers, giving them distinctive chemical and physical properties. Compared to conventional linear polymers, dendrimers, particularly PAMAM dendrimers, offer several advantages, including a well-defined structure, high monodispersed similar to proteins, a large number of terminal functional groups, branched inner cavities, ease of surface modification, and non-immunogenicity. These features make dendrimers highly beneficial in biomedicine, especially in cancer treatment. PAMAM dendrimers, in particular, are used as effective platforms for loading and delivering a wide range of therapeutic drugs and diagnostic agents, enhancing the transport of pharmaceuticals and genes for targeted therapy.

Polymeric micelles

Polymeric micelles are nanoscale structures composed of a hydrophilic shell surrounding a hydrophobic core with multiple internal compartments. These micelles self-assemble when the concentration exceeds the critical micelle concentration (CMC), typically ranging in size from 10 to 200 nm. Increasingly recognized as promising drug delivery systems, polymeric micelles offer several advantages over free drugs, including improved solubility, better

pharmacokinetics in vivo, and enhanced stability of the drug payload. Their size can be tailored to avoid rapid clearance via glomerular filtration, thereby extending their circulation time. Additionally, the properties of polymeric micelles facilitate efficient endosomal uptake and enhance the cellular localization of the drug-loaded micelles, improving their therapeutic effectiveness.

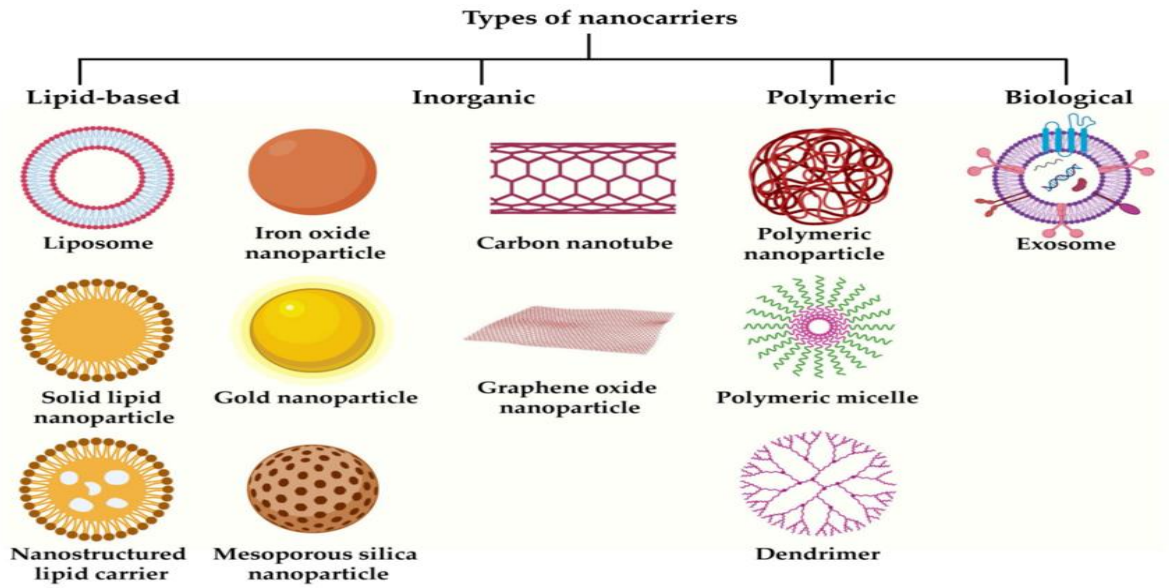

Figure 1: Various types of nanocarriers.

NANOSTRUCTURED LIPID CARRIER

Recently, lipid nanoparticles, particularly Solid Lipid Nanoparticles (SLNs) and Nanostructured Lipid Carriers (NLCs), have attracted significant attention in drug delivery research. SLNs are made from solid lipids, which create a crystalline structure with limited space for drugs, providing controlled release and protection from degradation. On the other hand, NLCs, an advanced form of lipid nanoparticles, combine both solid and liquid lipids. This unique structure improves drug loading capacity and stability, offering distinct advantages over SLNs. NLCs have shown promise in enhancing drug delivery, but they do face several challenges. For instance, some drugs, like quercetin, have low permeability and bioavailability, which can reduce their effectiveness despite the NLC's ability to improve solubility. Additionally, NLCs can become unstable over time, leading to phase separation or precipitation, which affects their consistency and clinical reliability. In dermatological applications, NLCs help reduce skin irritation, but they may still cause adverse reactions in some cases. In cancer treatments, NLCs improve drug targeting and minimize side effects compared to traditional methods, though their non-specific action could still impact healthy

cells. For wound healing, NLCs improve drug delivery to the affected areas, but external factors can interfere with the drug release process, reducing their overall effectiveness.

The NLC structure incorporates a blend of solid and liquid lipids, creating an irregular matrix that not only improves drug loading but also prevents drugs from leaching out during storage. This unique combination enhances stability by preventing solid lipids from recrystallizing, ensuring that the particle size remains consistent over time. The hybrid lipid matrix, with a particle size typically ranging from 10 to 500 nm, also allows for better control of drug release. NLCs provide many of the same benefits as SLNs, including reduced toxicity, biodegradability, and controlled release, without the need for organic solvents during production. NLCs have shown to be more effective than SLNs in terms of drug loading, entrapment efficiency, and skin penetration. Their solid matrix helps immobilize the drug, preventing aggregation, and making them ideal carriers for both hydrophilic and hydrophobic drugs. The ability of NLCs to bypass the first-pass metabolism and enhance lymphatic absorption ensures that more drug reaches the target site, improving therapeutic efficacy. Furthermore, the sustained release of the drug from the lipid matrix reduces the frequency of dosing, improving patient compliance. When applied topically in gel form, NLCs form a monolayer lipid film on the skin that enhances hydration, prevents trans-epidermal water loss (TEWL), and maintains moisture, further supporting their use in skin-related treatments.

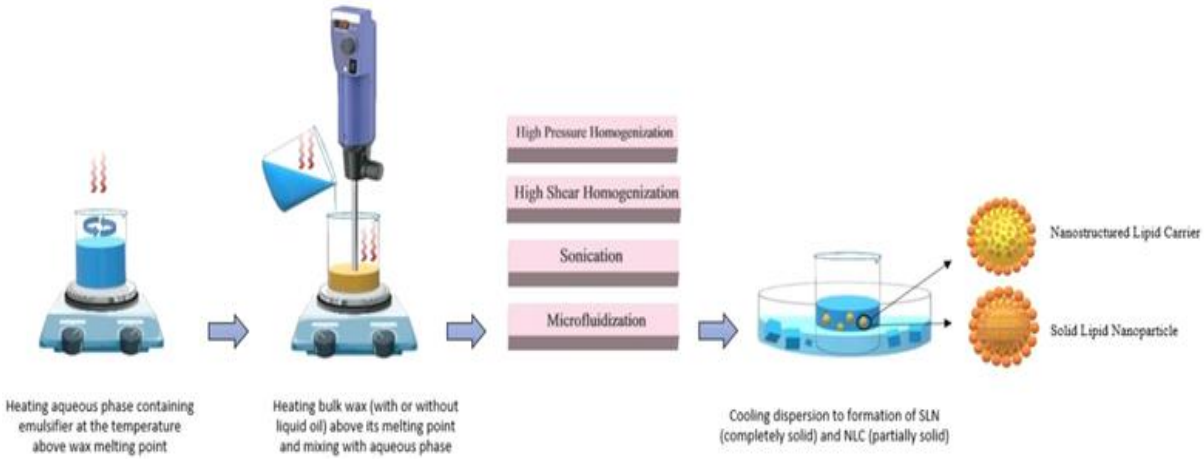

Figure 2: Key Steps in NLC Preparation Methods.

STRUCTURES AND PREPARATIONS OF NLCS

Materials for NLCs

Nanostructured Lipid Carriers (NLCs) are composed of essential ingredients such as lipids, water, and emulsifiers. The core structure of NLCs consists of both solid and liquid lipids.

Solid lipids typically used in NLCs include glyceryl behenate (Compritol® 888 ATO), glyceryl palmitostearate (Precirol® ATO 5), fatty acids (like stearic acid), triglycerides (e.g., tristearin), steroids (such as cholesterol), and waxes (e.g., cetyl palmitate). These lipids remain solid at room temperature but melt at higher temperatures (over 80°C) during the preparation process.

Liquid oils, generally derived from natural sources, are commonly used for NLCs. Medium-chain triglycerides like Miglyol® 812 are frequently employed due to their structural similarity to solid lipids such as Compritol®. Other oils used in NLC formulations include paraffin oil, 2-octyl dodecanol, propylene glycol dicaprylocaprate (Labrafac®), isopropyl myristate, and squalene. Fatty acids like oleic acid, linoleic acid, and decanoic acid are also included for their oily properties and their ability to act as penetration enhancers for topical drug delivery. These lipids are typically recognized as safe (GRAS) by European and American regulatory authorities. However, there is a need for novel, biocompatible oils that are cost-effective, non-irritating, and sterilizable for clinical use. Tocols, including Vitamin E (α-tocopherol), have also been explored for their stability, ease of production, and solubility in lipophilic drugs. Emulsifiers are essential for stabilizing lipid dispersions in NLCs. Most studies use hydrophilic emulsifiers such as Pluronic F68 (poloxamer 188), polysorbates (Tween), polyvinyl alcohol, and sodium deoxycholate. Lipophilic or amphiphilic emulsifiers like Span 80 and lecithin are used when necessary. Combining different emulsifiers can enhance the prevention of particle aggregation. Polyethylene glycol (PEG) is sometimes incorporated into the NLCs to help prevent their uptake by the reticuloendothelial system (RES) and extend the circulation time of the drugs. For NLCs to remain stable, proper preservation is crucial. However, preservatives can sometimes negatively affect the physical stability of lipid dispersions. Research by Obeidat et al. has demonstrated that Hydrolite® 5 is suitable for preserving coenzyme Q10-loaded NLCs, and light microscopy has been suggested as an efficient, cost-effective method for screening preservatives.

Table 1: The excipients for composing nanostructured lipid carriers (NLCs).

Ingredient	Material
Solid Lipids	Tristearin, stearic acid, cetyl palmitate, cholesterol, Precirol® ATO 5, Compritol® 888 ATO, Dynasan® 116, Dynasan® 118, Softisan® 154, Cutina® CP, Imwitor® 900 P, Geleol®, Gelot® 64, Emulcire® 61
Liquid lipids	Medium chain triglycerides, paraffin oil, 2-octyl dodecanol, oleic acid, squalene, isopropyl myristate, vitamin E, Miglyol® 812, Transcutol® HP, Labrafil Lipofile® WL 1349, Labrafac® PG, Lauroglycol® FCC, Capryol®

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Hydrophilic emulsifiers	Pluronic® F68 (poloxamer 188), Pluronic® F127 (poloxamer 407), Tween 20, Tween 40, Tween 80, polyvinyl alcohol, Solutol® HS15, trehalose, sodium deoxycholate, sodium glycocholate, sodium oleate, polyglycerol methyl glucose distearate
Lipophilic emulsifiers	Myverol® 18-04K, Span 20, Span 40, Span 60
Amphiphilic emulsifiers	Egg lecithin, soya lecithin, phosphatidylcholines, phosphatidylethanolamines, Gelucire®

Structures of NLCs

When Solid Lipid Nanoparticles (SLNs) are created using solid lipids, the resulting matrix tends to form a relatively perfect crystalline lattice, which restricts the available space for incorporating active ingredients. The inner core structure of SLNs typically has limited space for drugs. In contrast, Nanostructured Lipid Carriers (NLCs) are made using a blend of solid and liquid lipids, which distorts the formation of a perfect crystalline structure. This results in a matrix with imperfections, creating spaces where drug molecules can be accommodated in amorphous clusters. NLCs are often described as having oily droplets embedded within a solid lipid matrix. The particle morphology of NLCs is not necessarily spherical, and some researchers, such as Jores et al., suggest that the particulate structure consists of solid platelets with oil present between these platelets and the emulsifier layer.

Preparation Methods for NLCs

There are three primary methods for preparing NLCs: **hot homogenization**, **cold homogenization**, and **microemulsion**.

1. Hot Homogenization: This method is conducted at temperatures above the melting point of the lipids. First, the lipid phase (which includes both solid and liquid lipids along with lipophilic emulsifiers) and the aqueous phase (containing double-distilled water and hydrophilic emulsifiers) are prepared separately. Both phases are heated to a high temperature for a specified duration. The aqueous phase is then added to the lipid phase, and the mixture is homogenized using a high-shear homogenizer. To achieve smaller and more uniform particle sizes, the mixture may also be treated with a water-bath or probe-type sonicator. However, this high-temperature method can degrade heat-sensitive drugs. To address this, Hung et al. suggest reducing the heating temperature from 85°C to 60°C. This modification significantly reduces the degradation of sensitive drugs, such as vitamin E, compared to the conventional high-temperature method.

2. Cold Homogenization: In this method, the lipid melt is cooled, and the solid lipid is ground into microparticles. These microparticles are then dispersed in a cold emulsifier solution to form a pre-suspension. The suspension is subsequently homogenized at or below room temperature. The cavitation force during homogenization is sufficient to break the microparticles into NLCs. This approach avoids lipid melting, reducing the loss of hydrophilic drugs to the aqueous phase. However, the particle size may not always fall within the nanosized range due to the lack of high-temperature treatment.

3. Microemulsion: A microemulsion is formed when the melted lipids, emulsifiers, and water are mixed in a precise ratio. Adding this microemulsion to water causes the lipid phase to precipitate and form fine particles. This method is particularly suitable for large-scale production of lipid nanoparticles for the pharmaceutical industry. However, the resulting nanoparticle dispersion may be dilute, so it may need to be concentrated through ultrafiltration or lyophilization to achieve the desired product concentration.

These methods each have their advantages and limitations, but they offer different approaches to creating NLCs suitable for various pharmaceutical applications.

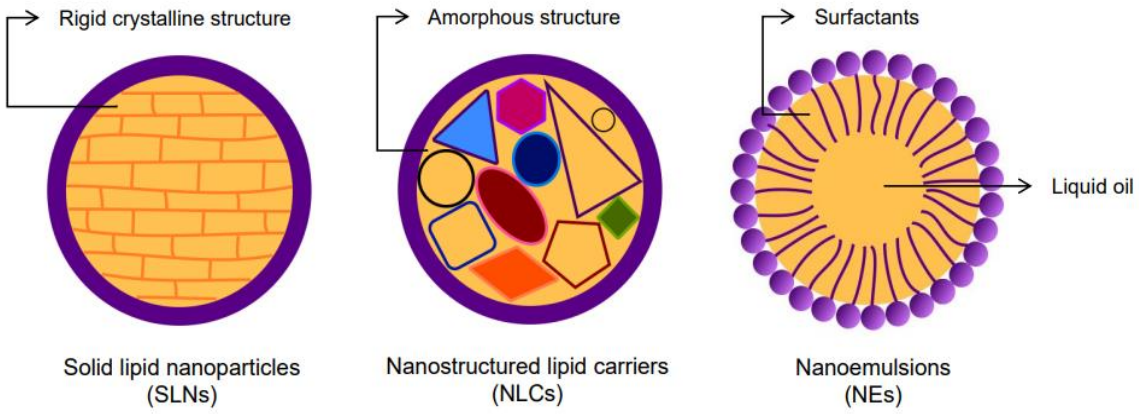

Fig. (3): Nanoparticulate structures of solid lipid nanoparticles (SLNs), nanostructured lipid carriers (NLCs), and oil-in-water nano emulsions (NES).

Physicochemical Characterization of NLCs

The physicochemical characterization of Nanostructured Lipid Carriers (NLCs) is crucial to ensure their quality control and stability. Both the physical and chemical properties of NLCs can be assessed. Common parameters for evaluating NLCs include particle size and zeta potential. Additionally, lipid nanoparticles are characterized using techniques like differential-scanning calorimetry (DSC), X-ray diffraction, nuclear magnetic resonance (NMR), and

Raman spectroscopy. The encapsulation efficiency and drug-release behaviour of NLCs are also important factors to evaluate their suitability as drug delivery systems.

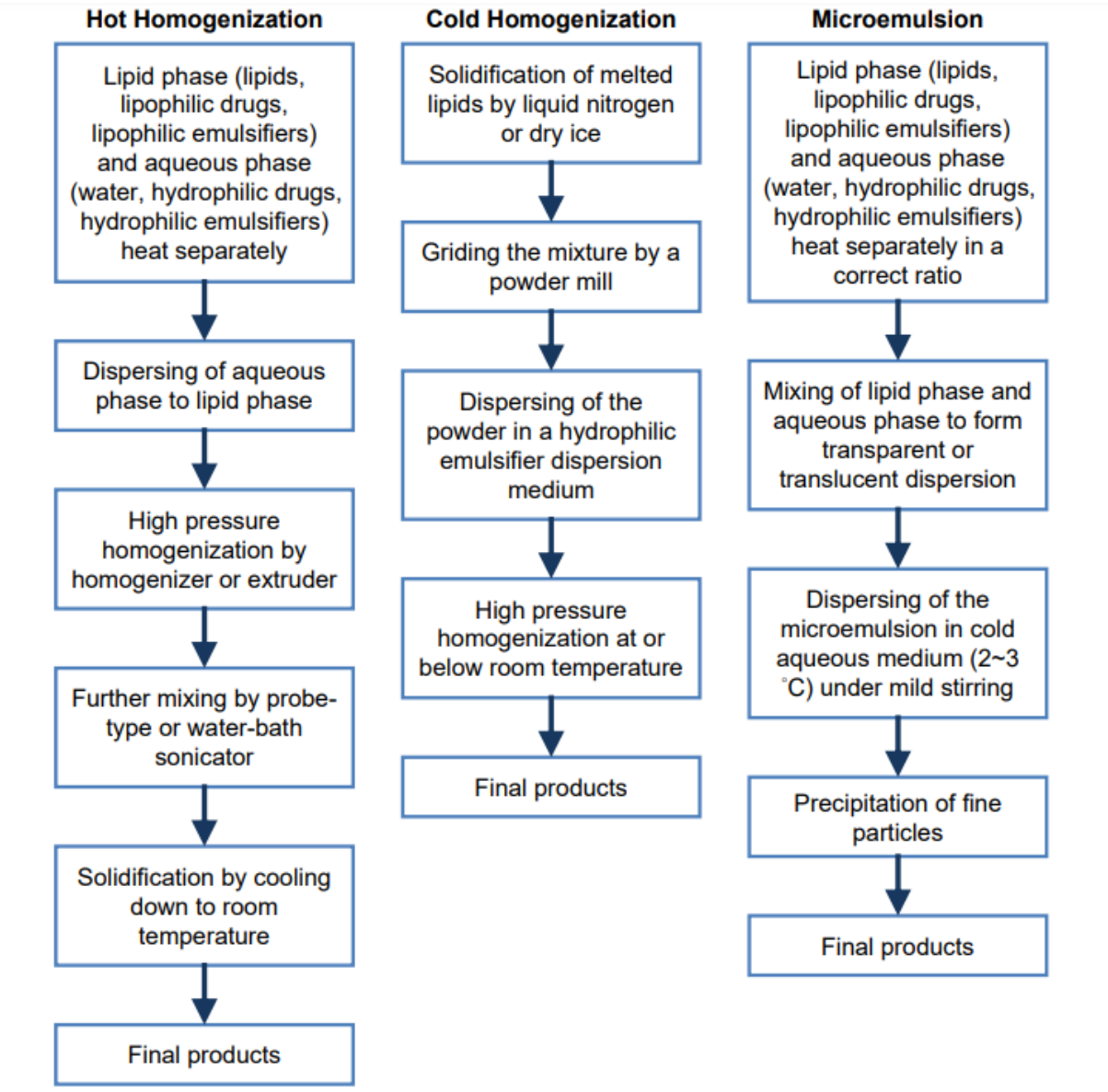

Fig. (4): Preparation procedures of nanostructured lipid carriers (NLCs): hot homogenization, cold homogenization, and microemulsion.

Particle Size

The most widely used techniques for measuring particle size are photon correlation spectroscopy (PCS), also known as dynamic light scattering, and laser diffraction. PCS measures the fluctuations in the intensity of scattered light caused by the movement of particles. It is capable of measuring particles within a size range from a few nanometres up to 3 micrometres. For larger particles, laser diffraction is used, which measures the angle of

diffraction in relation to the particle radius. The choice of lipid and emulsifier types, as well as their ratios, significantly influences the particle size. The inclusion of more emulsifiers can enhance emulsification, leading to a more rigid structure and a smaller particle size.

Zeta Potential

The zeta potential is a measure of the surface charge of NLCs and is an important factor in assessing their stability, dispersion, and aggregation behaviour. Particles with a higher surface charge are less likely to aggregate due to the electrostatic repulsion between them. For example, a positively charged surface can help NLCs cross the blood-brain barrier (BBB) by binding to the anionic sites in the paracellular area of the BBB. Zeta potential measurements are essential in formulation design to ensure that the desired surface charge, whether cationic or anionic, is achieved. A negative surface charge may be necessary for stabilizing the nanoparticulate systems during storage.

Electron Microscopy

Electron microscopy techniques, including scanning electron microscopy (SEM) and transmission electron microscopy (TEM), are commonly used to measure the size distribution and particulate radius of Nanostructured Lipid Carriers (NLCs). These methods also help in examining the shape and morphology of the particles. SEM works by reflecting electrons from the surface of the sample, while TEM transmits electrons through the sample. SEM is advantageous due to its high resolution and the simplicity of sample preparation, allowing the visualization of nanoparticles after processes such as freeze-drying or freeze-thawing.

Atomic Force Microscopy (AFM)

Atomic Force Microscopy (AFM) is particularly effective for analysing small-scale morphological and surface features. Unlike electron microscopy, AFM does not rely on photons or electrons but uses a sharp-tipped probe at the end of a cantilever. This probe interacts with the surface of the sample through repulsive or attractive forces at the atomic level. AFM offers significant advantages, including real-time, three-dimensional quantitative data acquisition, minimal sample preparation, flexibility under ambient conditions, and high magnification at the nanoscale. Despite the continued use of electron microscopy, AFM provides more flexibility and precision in observing the finer details of nanoparticulate structures.

Surface Tension

The surface tension of water at 20°C is typically 72.8 dynes/cm. The addition of lipids and emulsifiers significantly reduces the surface tension by disrupting the intermolecular forces within the system. As the concentration of emulsifiers increases, the surface tension continues to decrease due to the emulsification process. The surface tension of lipid nanoparticles is commonly measured using the Wilhelmy plate method, and contact angle measurements are another technique used to determine the surface tension of nanoparticulate systems.

Differential Scanning Calorimetry (DSC)

Differential Scanning Calorimetry (DSC) is a technique used to study the melting and recrystallization behaviour of solid lipids in both Solid Lipid Nanoparticles (SLNs) and NLCs. DSC works by measuring the heat flow associated with phase changes in the lipids, such as melting points and enthalpies. The degree of crystallinity in NLCs is calculated by comparing the enthalpy of the NLCs to the bulk lipid enthalpy, which is based on the total lipid weight. As the proportion of liquid lipids in the nanoparticles increases, the crystallinity of the particles decreases, which indicates that the liquid oil reduces the crystalline order and increases the disordered structure of the NLCs. This change creates more space within the particles, allowing for greater drug incorporation. DSC data also reveal the influence of emulsifiers and lipid composition on the melting point and enthalpy, suggesting the potential for preferential drug dissolution in either the solid or liquid lipid phases. This information is critical for understanding drug release behaviour and optimizing NLC formulations.

X-ray Diffraction

Both Differential Scanning Calorimetry (DSC) and X-ray diffraction are extensively used techniques to examine the structural properties of lipids. Lipid molecules, especially those containing long hydrocarbon chains, are known for their polymorphism. X-ray diffraction (XRD) can provide insights into the crystalline structure of NLCs by detecting the polymorphic form of the nanoparticles. The results from XRD are often used to verify the findings from DSC, especially in determining the crystallinity and the arrangement of lipid molecules within the nanoparticle structure. By analysing the X-ray scattering patterns, one can also determine the spacing between lipid lattices in both the long and short directions.

Parélectric Spectroscopy

Parélectric spectroscopy is based on the frequency-dependent behaviour of dipole density and mobility under the influence of an electromagnetic field. This technique is particularly useful

for understanding the structure and dynamics of Solid Lipid Nanoparticles (SLNs) and NLCs. Parélectric spectroscopy is a versatile tool as it provides valuable insights into the behaviour of liquid dispersions, and it can also be applied in medical diagnostics for testing biological materials.

Nuclear Magnetic Resonance (NMR)

Proton Nuclear Magnetic Resonance (NMR) spectroscopy is employed to study the mobility of the materials within the inner core of NLCs. The mobility of both solid and liquid lipids is reflected in the width of the NMR signals. A broader signal indicates restricted mobility due to stronger intermolecular interactions. The NMR spectra of NLCs exhibit wider line widths than those of the physical mixtures of the lipid components, suggesting stronger interactions between the liquid oil and solid lipid. This highlights the more immobilized nature of the NLCs compared to SLNs, which have fully crystallized cores.

Raman Spectroscopy

Raman spectroscopy detects molecular vibrations following the excitation of the sample by a laser beam. Water typically produces broad peaks around 3500 cm-1, while lipid order can be assessed by observing the bands associated with the stretching modes of methylene groups, typically found at 2840 cm-1 (symmetric stretching) and 2880 cm-1 (asymmetric stretching). These bands indicate a high degree of conformational order within the hydrocarbon chains of NLCs, which is a key feature of the lipid phase in NLCs.

Molecular Environment

The lipophilic fluorescent dye Nile Red can be used to probe the molecular environment within NLCs through fluorometric spectroscopy. Nile Red is a fluorescent compound that exhibits strong fluorescence in organic solvents or lipid environments. Its emission spectrum shifts depending on the polarity of the surrounding environment. In more non-polar lipid regions, the emission maximum is around 600 nm. As the polarity increases, such as in an aqueous environment or nanoparticulate shell, the emission shifts to shorter wavelengths and the fluorescence intensity decreases. In NLCs, Nile Red predominantly resides in the fluid lipid phase.

Drug Encapsulation Efficiency

Determining the drug-loading efficiency of NLCs is crucial because it directly impacts the release characteristics of the drug. Lipophilic drugs can either be uniformly distributed in the

lipid matrix, or preferentially incorporated into the core or shell of the nanoparticles. Hydrophilic drugs, on the other hand, tend to localize in the aqueous or interfacial phases. High drug-loading efficiency is typically achieved when the drug is highly soluble in the lipids, though this solubility may decrease upon cooling, especially in solid lipid phases. The encapsulation efficiency is determined by separating the internal and external phases using methods such as ultrafiltration, ultracentrifugation, gel filtration (e.g., using Sephadex), or dialysis. NLCs offer enhanced drug loading compared to SLNs, as the incorporation of liquid oils disrupts the crystalline order, allowing for greater drug entrapment in the disordered lipid matrix.

Drug Release

The controlled release of drugs from NLCs can lead to extended half-lives and protection from enzymatic degradation in systemic circulation. The release of drugs from NLCs depends on several factors, including production temperature, emulsifier composition, and the proportion of oil incorporated into the lipid matrix. Drugs in the outer shell or surface of the nanoparticles tend to release rapidly in a burst manner, whereas those within the core are released more slowly and continuously. The release behaviour can be attributed to the drug's partitioning between the lipid matrix and surrounding water, as well as the barrier properties of the interfacial membrane. In vitro drug release is commonly measured using dialysis techniques or Franz cells, and these profiles are important for predicting in vivo performance, considering factors such as enzymatic degradation.

Drug Delivery via NLCs

One of the most prominent applications of NLCs is as a drug delivery system. NLCs have been developed for various routes of administration, including parenteral injection, topical skin delivery, oral administration, ocular delivery, and pulmonary inhalation. Among these, injection and skin delivery are the most studied routes. NLCs are also emerging as carriers for gene delivery applications, offering potential for targeted and efficient delivery in genetic therapy.

PARENTERAL INJECTION

Brain Targeting

Parenteral administration of drugs via NLCs is a promising approach, particularly for brain targeting. Biodegradable materials in NLCs enhance drug stability and enable controlled drug release, offering prolonged therapeutic effects. One challenge in intravenous drug delivery is

the rapid uptake of nanoparticles by the mononuclear phagocyte system (MPS), which limits their ability to target areas beyond the reticuloendothelial system (RES). To overcome this issue, PEG (Polyethylene Glycol) and Pluronic F68 have been used to modify the surface of nanoparticles, preventing recognition by phagocytes and promoting the delivery of drugs to their intended target.

For example, NLCs containing Pluronic F68 have been successfully used to target the brain with bromocriptine, a drug used for treating Parkinson's disease. The slow release and specific targeting of bromocriptine through NLCs led to a longer therapeutic effect, as demonstrated in behavioural tests on Parkinson's disease models. NLCs were shown to extend the duration of the drug's action compared to control formulations, offering a more sustained treatment for motor disabilities in these models.

Similarly, apomorphine, another dopamine receptor agonist used to treat Parkinson's disease, has poor oral bioavailability and a short half-life. However, when formulated in apomorphine NLCs, its stability and brain-targeting ability were significantly improved, demonstrating better brain accumulation and prolonged action. In vivo imaging confirmed that apomorphine-loaded NLCs successfully targeted specific cerebral vessels.

Additionally, baicalein, a flavonoid with neuroprotective properties, showed enhanced brain delivery and stability when encapsulated in NLCs. The lipid nanoparticles significantly increased the plasma concentration and half-life of baicalein, leading to higher accumulation in brain regions such as the cerebral cortex, hippocampus, and thalamus, compared to the free drug. This reinforces the potential of NLCs in enhancing the delivery of neuroprotective agents for the treatment of ischemic brain injury and neurodegenerative diseases.

Tumor Targeting

NLCs are also gaining attention as drug carriers for cancer therapy, especially for improving the delivery and efficacy of anticancer drugs. The incorporation of NLCs can provide sustained release, improve the chemical stability of drugs, and enhance their cytotoxicity. For example, camptothecin and topotecan, when encapsulated in NLCs, showed increased cytotoxicity and enhanced uptake by melanoma and leukemia cells compared to free drug formulations.

Furthermore, the development of folate-conjugated NLCs represents a novel approach for tumor targeting. Folate receptors are overexpressed in many tumor cells, and the use of folic acid as a targeting ligand helps direct the NLCs specifically to cancer cells. One such formulation, containing docetaxel (an anticancer drug used for treating breast, lung, ovarian, and prostate cancers), demonstrated significant improvements in pharmacokinetics and tumor targeting. The docetaxel-loaded NLCs, when modified with a folate-polyethylene glycol copolymer, resulted in a higher area under the curve (AUC) and a longer half-life, significantly enhancing the drug's therapeutic potential.

Other formulations of docetaxel-loaded NLCs, consisting of biodegradable materials such as stearic acid, glyceryl monostearate, and soy lecithin, have been found to exhibit stronger cytotoxicity against cancer cells, such as A549 lung cancer cells. These formulations induced greater apoptosis and cell cycle arrest, and they showed improved efficacy in melanoma-bearing mice, with much lower toxicity compared to commercially available treatments.

Moreover, the combination of VEGFR-2 antibody-modified NLCs with docetaxel has proven effective for both tumor and vascular targeting. VEGFR-2, which is overexpressed in tumor cells and neovasculature, provides a unique target for this dual-targeting approach. In vivo studies revealed that docetaxel NLCs targeting both tumor cells and blood vessels resulted in significantly smaller tumor volumes in melanoma mice, demonstrating the enhanced antitumor efficacy of this treatment.

Another promising NLC formulation involves the dextran sulfate hybrid system for sustained release of mitoxantrone, which helps overcome multidrug resistance (MDR) in cancer cells. The introduction of dextran improved drug encapsulation and sustained release, leading to higher accumulation of the drug in resistant breast cancer cells. This system demonstrated increased circulation time and significantly enhanced cytotoxicity, effectively bypassing the efflux pumps responsible for drug resistance.

In summary, NLCs are a highly versatile and effective platform for brain and tumor targeting, offering benefits such as improved drug stability, controlled release, enhanced bioavailability, and targeted delivery. These properties make NLCs a promising strategy for both neurodegenerative diseases and cancer therapy.

ANTIHEPATOTOXIC INJECTION

Silybin, a polyphenol derived from Silymarin, is the key biologically active compound in milk thistle seeds known for its antihepatotoxic properties. While most Nanostructured Lipid Carriers (NLCs) aim to avoid the Reticuloendothelial System (RES) in organs like the liver and spleen, liver targeting is essential for silybin to exert its antihepatotoxic effect. Jia et al. developed NLCs to encapsulate silybin for intravenous delivery, which had an average size of 232 nm and a zeta potential of -21 mV. These NLCs resulted in a two-fold increase in the Area Under the Curve (AUC) compared to the control solution in rabbit studies. Additionally, the mean residence time of silybin in NLCs was significantly longer than in the solution (7.9 hours vs. 1.6 hours). Tissue distribution studies in mice showed that 34% of silybin was accumulated in the liver, and 22% in the spleen. Bifendate, a compound used to treat hepatitis with minimal side effects, has also been encapsulated in NLCs for improved drug payloads and liver targeting. Feng et al. reported that NLCs encapsulating bifendate showed higher liver uptake compared to the free drug, with particle sizes around 217 nm.

Analgesia

Buprenorphine is a promising drug for chronic pain and opioid dependence. However, it undergoes significant first-pass metabolism, limiting its effectiveness when taken orally. Consequently, it is only available as an injectable or sublingual tablet. Its half-life is relatively short, at 2.75 hours. Wang et al. used linseed oil and cetyl palmitate to form the core of buprenorphine NLCs, which had a mean diameter of 180-200 nm. Ester prodrugs of buprenorphine were also loaded into NLCs to provide sustained release. In vivo tests on rats showed that buprenorphine propionate in NLCs significantly extended analgesic latency to 10 hours, compared to 3 hours for the aqueous control. The NLCs showed negligible toxicity, indicating that combining prodrugs with NLCs can effectively enhance analgesia.

Anti-inflammation

Xu et al. aimed to improve the therapeutic efficacy of dexamethasone acetate for inflammation by encapsulating it in NLCs. The NLCs, prepared using the emulsification-ultrasound technique, had a uniform size of 178 nm and a zeta potential of -38 mV. The drug entrapment efficiency was 91%, and in vivo testing in rats with carrageenan-induced pleuritis showed that dexamethasone in NLCs had an average peak concentration of 7.6 μg/ml in pleural exudate, which was 8.3 times higher than that of the control. The NLC formulation

exhibited stronger anti-inflammatory activity than the free drug at the same dose, making NLCs an effective carrier for glucocorticoids to enhance inflammation treatment.

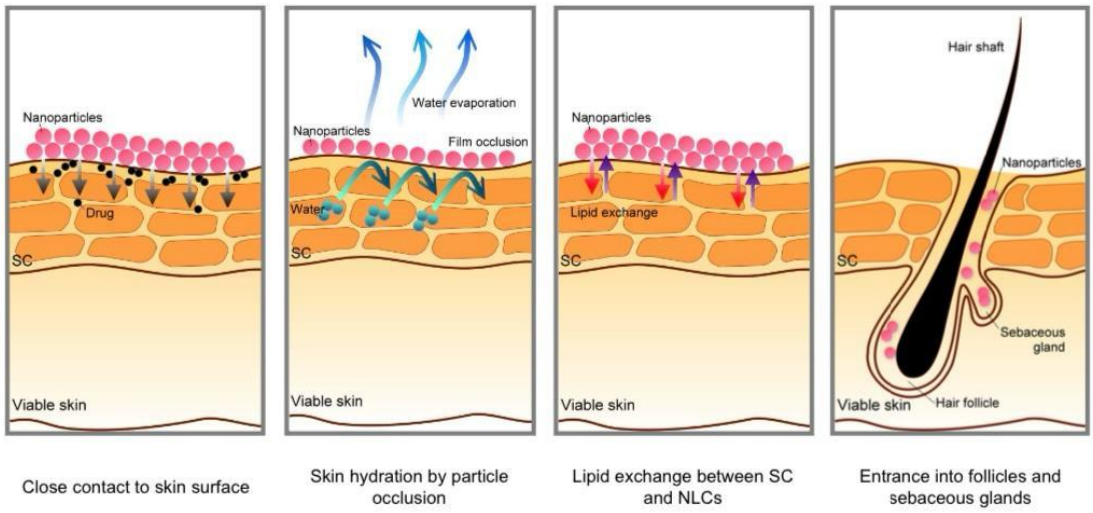

Fig. (5). Possible mechanisms for skin permeation enhancement of drugs or active ingredients from nanostructured lipid carriers (NLCs).

TOPICAL DELIVERY

Topical administration is one of the most explored routes for NLCs, as it can improve drug penetration through the skin. The stratum corneum (SC), however, is a significant barrier to drug delivery. Nanoparticulate systems enhance skin absorption and allow drug targeting to the skin or its deeper layers. The small size of nanoparticles allows for close contact with the SC, aiding in drug penetration. After the water evaporates from the applied nanoparticles, they form an adhesive layer on the skin, increasing hydration of the SC and widening gaps between corneocytes, which can further facilitate drug partitioning. While nanoparticles larger than 100 nm typically do not penetrate the SC, they can still provide benefits by enhancing the skin's lipid exchange. Lipid nanoparticles, in particular, have the potential to deliver drugs through hair follicles and sebaceous glands, which are lipid-rich environments that can trap these nanoparticles. This mechanism of enhanced skin permeation through NLCs is effective for both dermal drug delivery and reducing irritation, as the lipid matrix protects the skin from direct contact with the active drug. Acne vulgaris affects over 80% of individuals at some point in their lives, making it the most common skin disorder. Cyproterone acetate, a drug that reduces sebum production and acne lesions, can be more effectively delivered to hair follicles through nanoparticles. When cyproterone is incorporated into Nanostructured Lipid Carriers (NLCs), drug absorption through excised human skin

increases by 2-3 times. Another example is acitretin, a medication used for both acne and psoriasis treatment. When loaded into NLCs and combined with Carbopol 934 hydrogel, acitretin shows significantly higher deposition in human cadaver skin (81%) compared to plain gel (47%). Clinical studies also indicate that acitretin-loaded NLCs improve therapeutic response and reduce local side effects.

Photochemotherapy, involving psoralen and ultraviolet radiation (PUVA), is effective in clearing psoriasis. Three psoralen derivatives were loaded into Solid Lipid Nanoparticles (SLNs) and NLCs to evaluate their skin permeability. NLCs demonstrated enhanced permeation and controlled release, increasing psoralen flux 2.8 times more than conventional emulsions. Additionally, NLCs minimized the discrepancy in drug permeation between normal and psoriasis-like skin.

Anti-inflammatory drugs like flurbiprofen, ketoprofen, valdecoxib, celecoxib, and fluticasone are incorporated into NLCs for treating skin inflammation-related conditions. Flurbiprofen, a non-steroidal anti-inflammatory drug (NSAID) used for conditions such as gout and rheumatoid arthritis, has been investigated for skin permeation through rat skin. Studies show that flurbiprofen-loaded NLCs enhance permeation 4.5 times more than phosphate-buffered saline over 12 hours. Further research by González-Mira et al. showed improved delivery of flurbiprofen when loaded in NLCs composed of Compritol, with better particle size and crystallinity. NLCs were also shown to be non-irritant based on irritancy tests.

Additionally, a carrier system using cyclodextrin complexation combined with NLCs has been developed to enhance the efficacy of ketoprofen for treating arthritis and skin inflammation. The inclusion of cyclodextrin helps solubilize and stabilize the drug, leading to prolonged release and better skin absorption. When loaded into a xanthan hydrogel, this combination increased the ketoprofen permeation by two times in 6 hours, highlighting the synergistic effect of cyclodextrin and NLCs in promoting drug absorption.

The conventional valdecoxib formulation contains 56% alcohol, which can cause skin dryness. In contrast, Nanostructured Lipid Carriers (NLCs) show a burst release followed by a steady release of the drug, while the commercial formulation releases 100% of the drug within an hour. NLCs also exhibit prolonged anti-inflammatory activity, lasting up to 24 hours. A similar extended-release effect is observed for celecoxib, another COX-2 inhibitor, when formulated in NLCs. Fluticasone propionate, a glucocorticoid used to manage

inflammatory skin disorders such as atopic dermatitis and psoriasis, has been incorporated into NLCs to improve safety and reduce common adverse effects of topical corticoid treatments. The NLCs developed by Doktorovová et al. contain Precirol® and a mixture of PEG-containing medium-chain triglycerides, Tween 80, and soybean phosphatidylcholine as emulsifiers. These NLCs demonstrated a low-crystalline structure with high drug entrapment efficacy (97%) and stable particle size for up to 60 days.

Lidocaine, a local anaesthetic, is typically used for rapid onset and intermediate action with low systemic toxicity. NLCs and Solid Lipid Nanoparticles (SLNs) loaded with lidocaine have been formulated into hydrogels for topical application. In a study involving guinea pig skin, NLCs (73 nm) and SLNs (78 nm) showed more effective sustained release of lidocaine compared to the commercial product, Xylocaine®. In vivo tests revealed that NLCs and SLNs prolonged anaesthesia by 5-fold and 6-fold, respectively, compared to Xylocaine® gel. Puglia et al. also investigated the prolonged release of lidocaine and benzocaine from NLCs. Their study demonstrated that both drugs released in a sustained manner, producing a longer-lasting effect compared to a rapid, short-term effect seen with aqueous solutions.

Combining two drugs with different polarities into a single formulation can be challenging. However, by using NLCs with both lipid and aqueous phases, drugs like calcipotriol (a vitamin D3 analog for psoriasis) and methotrexate (used for reducing psoriasis symptoms) can be effectively incorporated. Lin et al. loaded calcipotriol in the lipid phase and methotrexate in the aqueous phase of NLCs. Their findings showed that NLCs enhanced skin permeation of methotrexate by 2.4 to 4.4 times compared to the aqueous solution, while calcipotriol flux was comparable to the control. This study suggests that NLCs can serve as an effective multidrug carrier, enhancing drug absorption while minimizing skin irritation.

In the cosmetic industry, NLCs and SLNs are used to deliver active ingredients, such as perfumes, sunscreen agents, and antioxidants, with extended release. These lipid nanoparticles are beneficial for blocking UV radiation due to their crystalline structure, which scatters UV light. Combining molecular sunscreens with SLNs enhances their sun-protective effects. NLCs also improve chemical stability, film formation, skin hydration, and bioavailability, while reducing skin irritation.

Nano Lipid Restore CLR® was the first NLC-based commercial cosmetic product, launched in 2006 by Chemisches Laboratorium Dr. Kurt Richter GmbH. It consists of carnauba wax

and black currant seed oil, designed as an excipient for cosmetics. Another product, NanoLipid Q 10 CLR®, contains coenzyme Q10, an endogenous antioxidant commonly used in anti-aging products. This formulation enhances coenzyme Q10 penetration into the skin, improving its effectiveness. An in vivo study showed that NanoRepair Q 10® (another product by Dr. Rimpler GmbH) has superior skin absorption compared to traditional oil-in-water emulsions. After 7 days of use, NanoRepair Q 10® significantly improved skin hydration, as confirmed by a Corneometer test. 74% of volunteers rated the increased skin hydration as very good or good. Dr. Rimpler GmbH also offers NanoVital®, a product that includes nano-sized titanium dioxide (TiO2) as a UV blocker to minimize photoaging, along with ursolic acid and oleanolic acid for their anti-inflammatory effects.

NLCs formulated with cetyl palmitate and varying amounts of caprylic/capric triacylglycerols were developed for the topical delivery of coenzyme Q10. These NLCs exhibited an average particle size of 180–240 nm with a narrow polydispersity index of 0.2, and nearly 100% entrapment efficiency. The in vitro release profile of coenzyme Q10 from the NLCs demonstrated a biphasic pattern, initially releasing the drug quickly, followed by sustained release. The rapid release in the first two hours is attributed to the enrichment of coenzyme Q10 in the nanoparticle shell. Junyaprasert et al. incorporated coenzyme Q10 NLCs into xanthan gum-based hydrogels for further investigation. Compared to nano emulsion-based hydrogels, the NLCs showed superior skin permeation due to their better skin-occlusion properties. These lipid nanoparticles maintained their nano-sized range and high entrapment efficiency (>90%) even after 12 months of storage at 40°C.

NLCs have also been used for the topical delivery of lutein, a potent antioxidant, blue light filter, and skin protectant against photodamage. Studies showed that NLCs can protect lutein from UV degradation, with only 6–8% lutein degradation observed in NLCs after UV irradiation at minimal erythema dose energy levels. In contrast, nano emulsions exhibited 14% degradation of lutein. The in vitro permeation study using pig ear skin showed negligible lutein penetration from NLCs, suggesting that lutein remained within the skin without systemic absorption. In comparison, nano emulsions delivered higher amounts of lutein to the receptor.

NLCs also serve as effective UV filters. Puglia et al. tested the percutaneous absorption and photostability of octyl-methoxycinnamate, a common UVB absorber, when loaded into NLCs. The results indicated that octyl-methoxycinnamate, when encapsulated in NLCs,

exhibited lower flux compared to Solid Lipid Nanoparticles (SLNs), meaning it stayed more effectively on the skin surface without penetrating. Moreover, NLCs demonstrated superior photostability, efficiently protecting the UV absorber from UV-induced degradation. Additionally, NLCs incorporating organic UV filters showed enhanced sun protection factor (SPF), achieving up to 45% higher SPF compared to conventional nano emulsions. These NLC formulations exhibited a synergistic effect with sunscreens, offering improved UV protection.

ORAL DELIVERY

There has been growing interest in using NLCs (Nanostructured Lipid Carriers) for oral drug delivery in recent years. These lipid-based carriers improve bioavailability and prolong plasma levels of drugs when administered orally. NLCs can protect drugs from the harsh conditions in the gastrointestinal tract and are especially effective for encapsulating lipophilic drugs that are otherwise insoluble. For example, repaglinide, an anti-diabetic medication with poor water solubility, suffers from low oral bioavailability and a short half-life. NLCs were developed to enhance its oral delivery. Date et al. created repaglinide-loaded NLCs using Gelucire 50/13, an amphiphilic lipid excipient that improves the aqueous solubility of lipophilic drugs. The study showed that NLCs significantly reduced blood glucose levels in rats, achieving approximately twice the reduction compared to conventional repaglinide tablets. In another study, NLCs composed of Precirol® and squalene were developed to enhance the bioavailability of lovastatin, a cholesterol-lowering agent. Over 70% of lovastatin molecules were successfully encapsulated in NLCs. The in vitro release showed a 60% reduction in the drug release rate, and oral bioavailability in rats improved from 4% to 24% with NLC delivery. NLCs also displayed more consistent bioavailability across subjects compared to the control solution. Real-time bioimaging confirmed the stability of NLCs in the gastric environment. Zhang et al. investigated NLCs as oral delivery systems for etoposide, a poorly water-soluble chemotherapeutic agent. They added PEG or DSPE-PEG to stabilize the NLCs and extend circulation time. The study found that smaller NLCs were more effective in crossing the intestinal wall. A pharmacokinetic study in rats showed that NLCs enhanced the bioavailability of etoposide by 1.8, 3.0, and 3.5 times for standard NLCs, PEG-NLCs, and DSPE-PEG-NLCs, respectively, compared to a control dispersion. The DSPE-PEG NLCs exhibited the highest cytotoxicity against lung carcinoma cells.

OCULAR DELIVERY

SLNs and NLCs offer several advantages for ocular drug delivery, including improved local tolerance, the ability to entrap lipophilic drugs, protection of labile compounds, and controlled release. SLNs have been used for ocular applications for decades, and more recent studies have explored NLCs as effective ocular delivery systems. Triamcinolone acetonide, a corticosteroid used for inflammatory and angiogenic ocular diseases, is typically administered via intravitreal injection. Araújo et al. encapsulated this drug in NLCs to increase its bioavailability when administered via ocular instillation. The resulting NLCs were nanometric (~200 nm), unimodal, and negatively charged, produced using high-pressure homogenization. The study showed minimal particulate aggregation after 6 months of storage at room temperature, with less than 1.5% backscattering observed. Strong fluorescence from a dye used to track the NLCs was detected on the anterior segment of the eye for up to 160 minutes after treatment, while no fluorescence was observed with the aqueous solution. These findings indicate that NLCs effectively deliver triamcinolone to the posterior segment of the eye through both corneal and non-corneal pathways.

Flurbiprofen-Loaded NLCs for Ocular Therapy

Flurbiprofen-loaded NLCs, using Compritol® or stearic acid as solid lipids, have been developed for anti-inflammatory ocular treatment. Both types of NLCs were incorporated into carbomer hydrogels to evaluate their ability to extend corneal residence time. The formulations exhibited plasticity with low or no thixotropic behaviour, making them suitable for ocular application. These NLCs sustained the release of flurbiprofen. In vitro tests using isolated rabbit corneas showed that NLCs enhanced drug penetration, with stearic acid-based NLCs demonstrating superior delivery. In vivo ocular tolerance in rabbits was confirmed using the Draize test.

Tian et al. developed surface-modified NLCs for ocular delivery of flurbiprofen using partially deacetylated water-soluble chitosan. These NLCs had an average particle size of approximately 80 nm, which minimized the potential for eye irritation. Trans corneal penetration tests with isolated rabbit corneas showed a significant increase in permeability. The chitosan-coated NLCs demonstrated a 1.4- to 1.8-fold higher permeability coefficient compared to plain NLCs and a phosphate solution. Precorneal retention studies in vivo indicated that the area under the curve (AUC) for chitosan-coated NLCs was 1.3 times higher

than plain NLCs and 2.4 times greater than the control, highlighting their potential for effective ocular drug delivery.

PULMONARY DELIVERY

NLCs offer multiple advantages for pulmonary drug delivery. Their small size and lipophilic nature enhance their bio adhesive properties, leading to prolonged residence time in the lungs. With a particle size under 500 nm, NLCs show increased diffusion mobility, improving lung deposition. Additionally, their controlled-release behaviour can extend therapeutic effects and reduce the need for frequent inhalations. Pardeike et al. explored NLCs for pulmonary applications by examining their storage stability and physicochemical properties before and after nebulization. Itraconazole, a broad-spectrum antifungal, was used as a model drug. The NLCs, composed of Precirol® and oleic acid, showed no particle growth when stored at room temperature or refrigerated. With an entrapment efficiency of 99%, the NLCs retained their drug-loading capacity over 6 months. The formulation displayed a burst release of itraconazole, and nebulizing it with either a jet stream or ultrasonic nebulizer did not affect particle size or entrapment efficiency, making it suitable for pulmonary use. In lung cancer treatment, celecoxib, known for its ability to inhibit carcinoma growth in combination with docetaxel, was encapsulated in NLCs and tested for lung deposition following nebulization in mice. The NLCs, with a particle size of 217 nm and an entrapment efficiency over 90%, released celecoxib in a controlled manner. Nebulized NLCs showed a 4-fold higher AUC in lung tissues compared to celecoxib solution. Additionally, NLCs exhibited slower systemic clearance (0.93 L/h) compared to the control (20.03 L/h), indicating their potential for prolonged therapeutic action.

GENE DELIVERY

The rise of gene therapy has prompted the development of new delivery systems for treating various diseases. Nonviral vectors, particularly lipid nanocarriers like liposomes and SLNs, have garnered attention due to their favourable safety profiles and effectiveness in delivering enzymes, proteins, nucleic acids, and DNA. A novel gene delivery system was created by modifying NLCs with acetylated polyethyleneimine (PEI), a polymer that can form complexes with anionic DNA, enabling efficient delivery to the cell nucleus via endosomal escape. In vitro studies showed that adding triolein to the NLCs enhanced transfection efficiency, likely by destabilizing the endosomal membrane. When compared to commercial

Lipofectamine® 2000, NLCs maintained transfection efficiency even in the presence of 10% serum, suggesting they are effective gene transfer vectors.

Small-interfering RNA (siRNA) has strong potential for cancer therapy, particularly for silencing oncogenic targets like survivin. However, their short action remains a clinical limitation. Xue and Wong demonstrated that NLCs could improve the duration of siRNA activity by controlling degradation in lysosomes. Tailored NLCs delivering survivin-siRNA extended the siRNA knockdown period to 9 days. In an in vivo tumor model, mice treated with docetaxel and siRNA-loaded NLCs showed a 65% to 55% reduction in tumor volume compared to the control and negative-siRNA groups. This innovative approach offers a longer administration cycle for siRNA therapy, potentially improving the clinical efficacy of cancer treatments.

PATENTS IN NLCS FOR DRUG DELIVERY

Table 2 provides a summary of recent patents related to NLCs for drug delivery applications. Notably, there are only a few patents in this field, with only one specifically using the term "nanostructured lipid carriers" in its title. The delivery of therapeutic compounds to cells often faces two main challenges: low drug selectivity and the restricted movement of compounds across complex cell membranes. Chen et al. have patented a novel particle delivery system that includes cationic lipids, microparticles, and nanoparticles, which are effective in delivering a range of molecules, including antibodies, hormones, proteins, vitamins, nucleic acids, and RNA, to cells. Keck and Muchow have developed NLCs for testosterone esters, particularly testosterone undecanoate. The formulation process involves incorporating testosterone undecanoate into a lipid matrix that is solid at body temperature, followed by mixing the drug dissolved in oil with a melted solid lipid (e.g., stearic acid) and dispersing the mixture in a hot surfactant solution. This method results in NLCs with enhanced oral bioavailability compared to existing oral formulations, and these nanonized formulations could also be suitable for dermal or nasal applications.

Table 2: The patents of NLCs for drug delivery application.

Publication number	Title	Inventors	Publication date
WO2011116963 A2	Lipid nanoparticle capsules	Viladot Petit V et al.	2011-09-29
US20110097392 A1	Antibody bound synthetic vesicle containing molecules for deliver to central and peripheral nervous system cells	Wang KK et al.	2011-04-28

US20110059157 A1	Anionic lipids and lipid nano-structures and methods of producing and using same	Awasthi V and Lagisetty P	2011-03-10
US20100247619 A1 WO2008000448 A3	Nanostructured lipid carriers containing riluzole and pharmaceutical formulations containing said particles	Bondi' ML et al.	2010-09-30
US20100047297 A1	Nanocrystals for use in topical cosmetic formulations and method of production	Petersen R	2010-02-25
US20090238878 A1	Solid nanoparticle formulation of water insoluble pharmaceutical substances with reduced Ostwald ripening	Singh CU	2009-09-24
EP2229936 A1	Nanonized testosterone formulations for improved bioavailability	Keck C and Muchow M	2009-03-09
US20080020058 A1	Lipid nanoparticles-based compositions and methods for the delivery of biologically active molecules	Chen T et al	2008-01-24

Singh filed a patent for a novel pharmaceutical composition that includes SLNs or NLCs dispersed in an aqueous medium containing poorly water-soluble substances like docetaxel, with reduced Ostwald ripening. An important feature of this patent is the use of proteins, particularly whey proteins such as β-lactoglobulin, α-lactalbumin, bovine serum albumin, and immunoglobulin, as emulsifiers. Petersen developed cosmetic formulations for topical use, incorporating nanocrystals of active ingredients to enhance bioactivity. These nanocrystals, with diameters ranging from 300 to 800 nm, can be included in formulations like liposomes, SLNs, and NLCs. Rutin, known for its antioxidant properties, was used as an active ingredient. The study revealed that these delivery systems are more stable during long-term storage and more electrolyte-stable compared to conventional formulations.

Bondi et al. incorporated riluzole, a neuroprotective drug, into NLCs for treating amyotrophic lateral sclerosis and multiple sclerosis. Using Compritol as the solid lipid, they achieved spherical nanoparticles with diameters under 100 nm. Their results showed that rats treated with riluzole-loaded NLCs exhibited delayed clinical signs of allergic encephalomyelitis compared to those receiving free riluzole. Additionally, the lipid nanoparticles crossed the blood-brain barrier (BBB) more efficiently than the free drug. Awasthi and Lagisetty patented lipid nanostructures for drug delivery, describing materials like 2-carboxyheptadecanoyl heptadecylamide (CHHDA), dipalmitoyl-tartarate derivatives, and cholesteryl hemisuccinate (CHEMS) for preparing the lipid matrix. Vitamin E was used as the liquid lipid to form the nanostructured matrix.

Wang et al. presented a novel process for delivering active agent cargo molecules into neuronal cells. The process involves incorporating the cargo into liposomes or NLCs and binding a biotinylated protein antibody to the nanoparticles, which targets receptors on neuronal cells in both the central and peripheral nervous systems (CNS and PNS).

Lastly, Petit et al. addressed the challenge of using lipids as matrices for formulating peptides/proteins, which are prone to proteolytic degradation in the gastrointestinal tract due to the hydrophobic nature of the matrix. They enhanced stability by applying a polymeric coating to SLNs or NLCs, providing additional protection for the peptides/proteins and preventing their diffusion in the lipid nanoparticles. This coating also improved the stabilization of the active ingredients compared to conventional lipid nanoparticles and enhanced the skin penetration capacity of the nanoparticles.

CURRENT & FUTURE DEVELOPMENTS

Choosing the right delivery vehicles is crucial for maximizing drug effectiveness while minimizing side effects. Recently, there has been increasing interest in novel nanocarriers, with NLCs standing out due to their excellent drug delivery potential and safety profile. This review highlights recent progress in drug delivery systems using NLCs. Besides intravenous administration, NLCs are also suitable for topical and oral routes. Lipid nanoparticles can address the limitations of conventional drug delivery vehicles. Given the urgent need for new treatments for conditions like cancer, neurodegenerative diseases, and inflammation, NLCs are expected to play a larger role in both basic research and clinical settings in the future. Numerous studies have focused on designing lipid nanoparticles to offer fewer side effects, longer half-lives, and better bioavailability compared to traditional carriers. However, only a few NLC formulations are currently used in clinical practice, with cosmetic products being the most common application. Clinical trials exploring NLCs for drug delivery are still limited, and more animal and clinical studies are needed to advance their use in therapeutic settings.

Although most ingredients used to create NLCs are biodegradable, the potential toxicity of nanoparticles remains a concern. Due to their small size and large surface area, nanomaterials may have more significant adverse effects than larger materials. The health risks associated with nanomaterials are still not fully understood, and there is limited information on the safety of nano-level substances. Intravenous injection and topical delivery are the primary methods for administering drugs using NLCs, but research into alternative routes, such as

gastrointestinal tract and blood-brain barrier (BBB) permeation, is expected to grow in the future. Another potential development is the combination of two active agents in a single NLC system. While the benefits of NLCs for drug delivery are evident, the mechanisms behind their enhanced efficacy remain unclear and require further investigation to optimize their therapeutic potential.

REFERENCES

1. Mehnert W, Mäder K. Solid lipid nanoparticles: production, characterization and applications. Adv Drug Deliv Rev, 2001; 47: 165- 96.
2. Carmona-Ribeiro AM. Biomimetic nanoparticles: preparation, characterization and biomedical applications. Int J Nanomed, 2010; 5: 249-59.
3. Sawant KK, Dodiya SS. Recent advances and patents on solid lipid nanoparticles. Recent Pat Drug Deliv Form, 2008; 2: 120-35.
4. Müller RH, Mäder K, Gohla S. Solid lipid nanoparticles (SLN) for controlled drug delivery–a review of the state of the art. Eur J Pharm Biopharm, 2000; 50: 161-77.
5. Jores K, Mehnert W, Mäder K. Physicochemical investigations on solid lipid nanoparticles and on oil-loaded solid lipid nanoparticles: a nuclear magnetic resonance and electron spin resonance study. Pharm Res, 2003; 20: 1274-83.
6. Müller RH, Radtke M, Wissing SA. Nanostructured lipid matrices for improved microencapsulation of drugs. Int J Pharm, 2002; 242: 121-8.
7. Jenning V, Thünemann AF, Gohla SH. Characterisation of a novel solid lipid nanoparticle carrier system based on binary mixtures of liquid and solid lipids. Int J Pharm, 2000; 199: 167-77.
8. Constantinides PP, Tustian A, Kessler DR. Tocol emulsions for drug solubilization and parenteral delivery. Adv Drug Deliv Rev, 2004; 56: 1243-55.
9. Tsai MJ, Wu PC, Huang YB, et al. Baicalein loaded in tocol nanostructured lipid carriers (tocol NLCs) for enhanced stability and brain targeting. Int J Pharm, 2012; 423: 461-70.
10. Averina ES, Seewald G, Müller RH, Radnaeva LD, Popov DV. Nanostructured lipid carriers (NLC) on the basis of Siberian pine (Pinus sibirica) seed oil. Pharmazie, 2010; 65: 25-31.
11. Averina ES, Müller RH, Popov DV, Radnaeva LD. Physical and chemical stability of nanostructured lipid carriers (NLC) based on natural lipids from Baikal region (Siberia, Russia). Pharmazie, 2011; 66: 348-56.

12. Schäfer-Korting M, Mehnert W, Korting HC. Lipid nanoparticles for improved topical application of drugs for skin diseases. Adv Drug Deliv Rev, 2007; 59: 427-43.
13. Rosenblatt KM, Bunjes H. Poly(vinyl alcohol) as emulsifier stabilizes solid triglyceride drug carrier nanoparticles in the - modification. Mol Pharm, 2009; 6: 105-20.
14. Gu X, Zhang W, Liu J, et al. Preparation and characterization of a lovastatin-loaded protein-free nanostructured lipid carrier resembling high-density lipoprotein and evaluation of its targeting to foam cells. AAPS PharmSciTech, 2011; 12: 1200-08.
15. Zhang X, Pan W, Gan L, et al. Preparation of a dispersible Pegylate nanostructured lipid carriers (NLC) loaded with 10- hydroxycamptothecin by spray-drying. Chem Pharm Bull, 2008; 56: 1645-50.
16. Obeidat WM, Schwabe K, Müller RH, Keck CM. Preservation of nanostructured lipid carriers (NLC). Eur J Pharm Biopharm, 2010; 76: 56-67.
17. Jores K, Haberland A, Wartewig S, Mäder K, Mehnert W. Solid lipid nanoparticles (SLN) and oil-loaded SLN studied by spectrofluorometry and Raman spectroscopy. Pharm Res, 2005; 22: 1887- 97.
18. Hung LC, Basri M, Tejo BA, et al. An improved method for the preparations of nanostructured lipid carriers containing heatsensitive bioactives. Colloids Surf B: Biointerf, 2011; 87: 180-6.
19. Joshi M, Patravale V. Formulation and evaluation of nanostructured lipid carrier (NLC)-based gel of valdecoxib. Drug Dev Ind Pharm, 2006; 32: 911-8.
20. Das S, Chaudhury A. Recent Advances in Lipid Nanoparticle Formulations with solid matrix for oral drug delivery. AAPS PharmSciTech, 2011; 12: 62-76.
21. Parveen S, Sahoo SK. Polymeric nanoparticles for cancer therapy. J Drug Target, 2008; 16: 108-23.
22. Hwang TL, Lin YK, Chi CH, Huang TH, Fang JY. Development and evaluation of perfluorocarbon nanobubbles for apomorphine delivery. J Pharm Sci, 2009; 98: 3735-47.
23. Sitterberg J, Özcetin A, Ehrhardt C, Bakowsky U. Utilising atomic force microscopy for the characterisation of nanoscale drug delivery systems. Eur J Pharm Biopharm, 2010; 74: 2-13.
24. Forny L, Saleh K, Denoyel R, Pezron I. Contact angle assessment of hydrophobic silica nanoparticles related to the mechanisms of dry water formation. Langmuir, 2010; 26: 2333-8.
25. Hu FQ, Jiang SP, Du YZ, et al. Preparation and characteristics of monostearin nanostructured lipid carriers. Int J Pharm, 2006; 314: 83-9.

26. Castelli F, Puglia C, Sarpietro MG, Rizza L, Bonina F. Characterization of indomethacin-loaded lipid nanoparticles by differential scanning calorimetry. Int J Pharm, 2005; 304: 231-8.
27. Sato K. Crystallization behaviour of fats and lipids–a review. Chem Eng Sci, 2001; 5: 2255-65.
28. Teeranachaideekul V, Müller RH, Junyaprasert VB. Encapsulation of ascorbyl palmitate in nanostructured lipid carriers (NLC)–effects of formulation parameters on physicochemical stability. Int J Pharm, 2007; 340: 198-206.
29. Sivaramakrishnan R, Nakamura C, Mehnert W, et al. Glucocorticoid entrapment into lipid carriers–characterization by parelectric spectroscopy and influence on dermal uptake. J Control Release, 2004; 97: 493-502.
30. Wissing SA, Müller RH, Manthei L, Mayer C. Structural characterization of Q10-loaded solid lipid nanoparticles by NMR spectroscopy. Pharm Res, 2004; 21: 400-5.
31. Schubert MA, Harms M, Müller-Goymann CC. Structural investigations on lipid nanoparticles containing high amounts of lecithin. Eur J Pharm Biopharm, 2006; 27: 226-36.
32. Saupe A, Gordon KC, Rades T. Structural investigations on nanoemulsions, solid lipid nanoparticles and nanostructured lipid carriers by cryo-field emission scanning electron microscopy and Raman spectroscopy. Int J Pharm, 2006; 314: 56-62.
33. Lombardi Borgia S, Regehly M, Sivaramakrishnan R, et al. Lipid nanoparticles for skin penetration enhancement–correlation to drug localization within the particle matrix as determined by fluorescence and parelectric spectroscopy. J Control Release, 2005; 110: 151-63.
34. Joshi M, Patravale V. Nanostructured lipid carrier (NLC) based gel of celecoxib. Int J Pharm, 2008; 346: 124-32.
35. Yuan H, Wang LL, Du YZ, et al. Preparation and characteristics of nanostructured lipid carriers for control-releasing progesterone by melt-emulsification. Colloids Surf B: Biointerf, 2007; 60: 174-9.
36. Marcato PD, Durán N. New aspects of nanoparmaceutical delivery systems. J Nanosci Nanotechnol, 2008; 8: 2216-29.
37. Joshi MD, Müller RH. Lipid nanoparticles for parenteral delivery of actives. Eur J Pharm Biopharm, 2009; 71: 161-72.
38. Esposito E, Mariani P, Ravani L, et al. Nanoparticulate lipid dispersions for bromocriptine delivery: characterization and in vivo study. Eur J Pharm Biopharm, 2012; 80: 306-14.

39. Chattopadhyay N, Zastre J, Wong HL, Wu XY, Bendayan R. Solid lipid nanoparticles enhance the delivery of the HIV protease inhibitor, atazanavir, by a human brain endothelial cell line. Pharm Res, 2008; 25: 2262-71.
40. Tomlinson CL, Stowe R, Patel S, et al. Systematic review of levodopa dose equivalency reporting in Parkinson's disease. Movement Disord, 2010; 25: 2649-53.
41. Subramony JA. Apomorphine in dopaminergic therapy. Mol Pharm, 2006; 3: 380-5.
42. Hsu SH, Wen CJ, Al-Suwayeh SA, et al. Physicochemical characterization and in vivo bioluminescence imaging of nanostructured lipid carriers (NLCs) for targeting the brain: apomorphine as a model drug. Nanotechnology, 2010; 21: 405101.
43. Liu C, Wu J, Xu K, et al. Neuroprotection by baicalein in ischemic brain injury involves PTEN/AKT pathway. J Neurochem, 2010; 112: 1500-12.
44. Mu X, He GR, Yuan X, Li XX, Du GH. Baicalein protects the brain against neuron impairments induced by MPTP in C57BL/6 mice. Pharmacol Biochem Behav, 2011; 98: 286-91.
45. Tsai MJ, Wu PC, Huang YB, et al. Baicalein loaded in tocol nanostructured lipid carriers (tocol NLCs) for enhanced stability and brain targeting. Int J Pharm, 2012; 423: 461-70.
46. Huang ZR, Hua SC, Yang YL, Fang JY. Development and evaluation of lipid nanoparticles for camptothecin delivery: a comparison of solid lipid nanoparticles, nanostructured lipid carriers, and lipid emulsion. Acta Pharmacol Sin, 2008; 29: 1094-102.
47. Souza LG, Silva EJ, Martins ALL, et al. Development of topotecan loaded lipid nanoparticles for chemical stabilization and prolonged release. Eur J Pharm Biopharm, 2011; 79: 189-96.
48. Zhao X, Zhao Y, Geng L, et al. Pharmacokinetics and tissue distribution of docetaxel by liquid chromatography-mass spectrometry: evaluation of folate receptor-targeting amphiphilic copolymer modified nanostructured lipid carrier. J Chromatogr B., 2011; 879: 3721-27.
49. Montero A, Fossella F, Hortobagyi G, Valero V. Docetaxel for treatment of solid tumours: a systematic review of clinical data. Lancet Oncol, 2005; 6: 229-39.
50. Liu D, Liu Z, Wang L, Zhang C, Zhang N. Nanostructured lipid carriers as novel carrier for parenteral delivery of dicetaxel. Colloids Surf B: Biointerf, 2011; 85: 262-9.
51. Liu D, Liu F, Liu Z, Wang L, Zhang N. Tumor specific delivery and therapy by double-targeted nanostructured lipid carriers with anti-VEGFR-2 antibody. Mol Pharm, 2011; 8: 2291-301.

52. Zhang P, Ling G, Pan X, et al. Novel nanostructured lipid-dextran sulfate hybrid carriers overcome tumor multidrug resistance of mitoxantrone hydrochloride. Nanomed-Nanotechnol Biol Med, 2012; 8: 185-93.
53. Gaák R, Walterová D, K en V. Silybin and silymarin–new and emerging applications in medicine. Curr Med Chem, 2007; 14: 315- 38.
54. Jia L, Zhang D, Li Z, et al. Nanostructured lipid carriers for parenteral delivery of silybin: biodistribution and pharmacokinetic studies. Colloids Surf B: Biointerf, 2010; 80: 213-8.
55. Pan SY, Yang R, Dong H, Yu ZL, Ko KM. Bifendate treatment attenuates hepatic steatosis in cholesterol/bile salt- and high-fat diet-induced hypercholesterolemia in mice. Eur J Pharmacol, 2006; 552: 170-5.
56. Feng F, Zheng D, Zhang D, Duan C, Wang Y. Preparation, characterization, and biodistribution of nanostructured lipid carriers for parenteral delivery of bifendate. J Microencapsul, 2011; 28: 280-5.
57. Ohtani M. Basic pharmacology of buprenorphine. Eur J Pain, 2007; Suppl 1: 69-73.
58. Escher M, Daali Y, Chabert J, et al. Pharmacokinetic and pharmacodynamic properties of buprenorphine after a single intravenous administration in healthy volunteers: a randomized, double-blind, placebo-controlled, crossover study. Clin Ther, 2007; 29: 1620-31.
59. Wang JJ, Liu KS, Sung KC, Tsai CY, Fang JY. Lipid nanoparticles with different oil/fatty ester ratios as carriers of buprenorphine and its prodrugs for injection. Eur J Pharm Sci, 2009; 38: 138-46.
60. Xu X, Zhao C, Yang H, et al. Anti-inflammatory activity of injectable dexamethasone acetate-loaded nanostructured lipid carriers. Drug Deliv, 2011; 18: 485-92.
61. Souto EB, Almeida AJ, Müller RH. Lipid nanoparticles (SLN®, NLC®) for cutaneous drug delivery: structure, protection and skin effects. J Biomed Nanotechnol, 2007; 3: 317-31.
62. Baroli B. Penetration of nanoparticles and nanomaterials in the skin: fiction or reality? J Pharm Sci, 2009; 99: 21-50.
63. Cevc G. Lipid vesicles and other colloids as drug carriers on the skin. Adv Drug Deliv Rev, 2004; 56: 675-711.
64. Müller RH, Petersen RD, Hommoss A, Pardeike J. Nanostructured lipid carriers (NLC) in cosmetic dermal products. Adv Drug Deliv Rev, 2007; 59: 522-30.
65. Chen H, Chang X, Du D, et al. Podophyllotoxin-loaded solid lipid nanoparticles for epidermal targeting. J Control Release, 2006; 110: 296-306.

66. Knorr F, Lademann J, Patzelt A, et al. Follicular transport route– research progress and future perspectives. Eur J Pharm Biopharm, 2009; 71: 173-80.
67. Nakatsuji T, Kao MC, Fang JY, et al. Antimicrobial property of lauric acid against Propionibacterium acnes: its therapeutic potential for inflammatory acne vulgaris. J Invest Dermatol, 2009; 129: 2480-8.
68. tecová J, Mehnert W, Blaschke T, et al. Cyproterone acetate loading to lipid nanoparticles for topical acne treatment: particle characterisation and skin uptake. Pharm Res, 2007; 24: 991-1000.
69. Lee CS, Koo J. A review of acitrecin, a systemic retinoid for the treatment of psoriasis. Expert Opin Pharmaco ther, 2005; 6: 1725- 34.
70. Agrawal Y, Petkar KC, Sawant KK. Development, evaluation and clinical studies of acitretin loaded nanostructured lipid carriers for topical treatment of psoriasis. Int J Pharm, 2010; 401: 93-102.
71. Stern RS. Psoralen and ultraviolet A light therapy for psoriasis. N Eng J Med, 2007; 357: 682-90.
72. Fang JY, Fang CL, Liu CH, Su YH. Lipid nanoparticles as vehicles for topical psoralen delivery: solid lipid nanoparticles (SLN) versus nanostructured lipid carriers (NLC). Eur J Pharm Biopharm, 2008; 70: 633-40.
73. Han F, Li S, Yin R, Shi X, Jia Q. Investigation of nanostructured lipid carriers for transdermal delivery of flurbiprofen. Drug Dev Ind Pharm, 2008; 34: 453-8.
74. González-Mira E, Nikoli S, García ML, et al. Potential use of nanostructured lipid carriers for topical delivery of flurbiprofen. J Pharm Sci, 2011; 100: 242-51.
75. Cirri M, Bragagni M, Mennini N, Mura P. Development of a new delivery system consisting in “drug – in cyclodextrin – in nanos-tructured lipid carriers” for ketoprofen topical delivery. Eur J Pharm Biopharm, 2012; 80: 46-53.
76. Doktorovová S, Araújo J, Garcia ML, Rakovsk E, Souto EB. Formulating fluticasone propionate in novel PEG-containing nanostructured lipid carriers (PEG-NLC). Colloids Surf B: Biointerf, 2010; 75: 538-42.
77. Pathak P, Nagarsenker M. Formulation and evaluation of lidocaine lipid nanosystems for dermal delivery. AAPS PharmSciTech, 2009; 10: 985-92.
78. Puglia C, Sarpietro MG, Bonina F, et al. Development, characterization, and in vitro and in vivo evaluation of benzocaine- and lidocaine-loaded nanostructured lipid carriers. J Pharm Sci, 2011; 100: 1892-9.

79. Fogh K, Kragballe K. New vitamin D analogs in psoriasis. Curr Drug Target Inflamm Allergy, 2004; 3: 199-204.
80. Lin YK, Huang ZR, Zhuo RZ, Fang JY. Combination of calcipotriol and methotrexate in nanostructured lipid carriers for topical delivery. Int J Nanomed, 2010; 5: 117-28.
81. Graf J. Antioxidants and skin care: the essentials. Plast Reconstr Surg, 2010; 125: 378-83.
82. Pardeike J, Schwabe K, Müller RH. Influence of nanostructured lipid carriers (NLC) on the physical properties of the Cutanova Nanorepair Q10 cream and the in vivo skin hydration effect. Int J Pharm, 2010; 396: 166-73.
83. Teeranachaideekul V, Souto EB, Junyaprasert VB, Müller RH. Cetyl palmitate-based NLC for topical delivery of coenzyme Q10– development, physicochemical characterization and in vitro release studies. Eur J Pharm Biopharm, 2007; 67: 141-8.
84. Junyaprasert VB, Teeranachaideekul V, Souto EB, Boonme P, Müller RH. Q10-loaded NLC versus nanoemulsions: stability, rheology and in vitro skin permeation. Int J Pharm, 2009; 377: 207- 14.
85. Roberts RL, Green J, Lewis B. Lutein and zeaxanthin in eye and skin health. Clin Dermatol, 2009; 27: 195-201.
86. Mitri K, Shegokar R, Gohla S, Anselmi C, Müller RH. Lipid nanocarriers for dermal delivery of lutein: preparation, characterization, stability and performance. Int J Pharm, 2011; 414: 267-75.
87. Puglia C, Bonina F, Rizza L, et al. Lipid nanoparticles as carrier for octyl-methoxycinnamate: in vitro percutaneous absorption and photostability studies. J Pharm Sci, 2012; 101: 301-11.
88. NikoliS, Keck CM, Anselmi C, Müller RH. Skin photoprotection improvement: synergistic interaction between lipid nanoparticles and organic UV filters. Int J Pharm, 2011; 414: 276-84.
89. Jain SK, Agrawal GP, Jain NK. A novel calcium silicate based microspheres of repaglinide: in vivo investigations. J Control Release, 2006; 113: 111-6.
90. Date AA, Vador N, Jagtap A, Nagarsenker MS. Lipid nanocarriers (GeluPearl) containing amphiphilic lipid Gelucire 50/13 as a novel stabilizer: fabrication, characterization and evaluation for oral drug delivery. Nanotechnology, 2011; 22: 275102.
91. Qi S, Marchaud D, Craig DQ. An investigation into the mechanism of dissolution rate enhancement of poorly water-soluble drugs from spray chilled gelucire 50/13 microspheres. J Pharm Sci, 2010; 99: 262-74.

92. Chen CC, Tsai TH, Huang ZR, Fang JY. Effects of lipophilic emulsifiers on the oral administration of lovastatin from nanostructured lipid carriers: physicochemical characterization and pharmacokinetics. Eur J Pharm Biopharm, 2010; 74: 474-82.
93. Zhang T, Chen J, Zhang Y, Shen Q, Pan W. Characterization and evaluation of nanostructured lipid carrier as a vehicle for oral delivery of etoposide. Eur J Pharm Sci, 2011; 43: 174-9.
94. Souto EB, Doktorovova S, Gonzalez-Mira E, Egea MA, García ML. Feasibility of lipid nanoparticles for ocular delivery of antiinflammatory drugs. Curr Eye Res, 2010; 35: 537-52.
95. Cavalli R, Gasco MR, Chetoni P, Burgalassi S, Saettone MF. Solid lipid nanoparticles (SLN) as ocular delivery system for tobramycin. Int J Pharm, 2002; 238: 241-5.
96. Attama AA, Reichl S, Müller-Goymann CC. Diclofenac sodium delivery to the eye: in vitro evaluation of novel solid lipid nanoparticle formulation using human cornea construct. Int J Pharm, 2008; 355: 307-13.
97. Gokce EH, Sandri G, Bonferoni MC, et al. Cyclosporine A loaded SLNs: Evaluation of cellular uptake and corneal cytotoxicity. Int J Pharm, 2008; 364: 76-86.
98. Araújo J, Nikolic S, Egea MA, Souto EB, García ML. Nanostructured lipid carriers for triamcinolone acetonide delivery to the posterior segment of the eye. Colloids Surf B: Biointerf, 2011; 88: 150- 7.
99. Gonzalez-Mira E, Nikoli S, Calpena AC, et al. Improved and safe transcorneal delivery of flurbiprofen by NLC and NLC-based hydrogels. J Pharm Sci 2012; 101: 707-25.
100. Tian B, Luo Q, Song S, et al. Novel surface-modified nanostructured lipid carriers with partially deacetylated water-soluble chitosan for efficient ocular delivery. J Pharm Sci, 2012; 101: 1040-9.
101. Jaques PA, Kim CS. Measurement of total lung deposition of inhaled ultrafine particles in healthy men and women. Inhal Toxicol, 2000; 12: 715-31.
102. Pardeike J, Weber S, Haber T, et al. Development of an itraconazole-loaded nanostructured lipid carrier (NLC) formulation for pulmonary application. Int J Pharm, 2011; 419: 329-38.
103. Hida T, Kozaki KI, Muramatsu H, et al. Cyclooxygenase-2 inhibitor induces apoptosis and enhances cytotoxicity of various anticancer agents in non-small cell lung cancer cell lines. Clin Cancer Res, 2000; 6: 2006-11.

104. Patlolla RR, Chougule M, Patel AR, et al. Formulation, characterization and pulmonary deposition of nebulized celecoxib encapsulated nanostructured lipid carriers. J Control Release, 2010; 144: 233-41.
105. del Pozo-Rodríguez A, Solinís MA, Gascón AR, Pedraz JL. Shortand long-term stability study of lyophilized solid lipid nanoparticles for gene therapy. Eur J Pharm Biopharm, 2009; 71: 181-9.
106. Zhang Z, Sha X, Shen A, et al. Polycation nanostructured lipid carrier, a novel nonviral vector constructed with triolein for efficient gene delivery. Biochem Biophys Res Commun, 2008; 370: 478-82.
107. Thomas M, Klibanov AM. Enhancing polyethylenimine's delivery of plasmid DNA into mammalian cells. Proc Natl Acad Sci USA, 2002; 99: 14640-5.
108. Pirollo KF, Chang EH. Targeted delivery of small interfering RNA: approaching effective cancer therapies. Cancer Res, 2008; 68: 1247- 50.
109. Xue HY, Wong HL. Tailoring nanostructured solid-lipid carriers for time-controlled intracellular siRNA kinetics to sustain RNAimediated chemosensitization. Biomaterials, 2011; 32: 2662-2672.
110. Chen T, Vargeese C, Vagle K, Wang W, Zhang Y. Lipid nanoparticles based compositions and methods for the delivery of biologically active molecules. US20080020058, 2008.
111. Keck C, Muchow M. Nanonized testosterone formulations for improved bioavailability. EP2229936, 2009.
112. Singh CU. Solid nanoparticle formulation of water insoluble pharmaceutical substances with reduced Ostwald ripening. US20090238878 A1, 2009.
113. Petersen R. Nanocrystals for use in topical cosmetic formulations and method of production thereof. US20100047297, 2010.
114. Bondi' ML, Giammona G, Craparo EF, Drago F. Nanostructured lipid carriers containing riluzole and pharmaceutical formulations containing said particles. US20100247619, 2010.
115. Awasthi V, Lagisetty P. Anionic lipids and lipid nano-structures and methods of producing and using same. US20110059157, 2011.
116. Wang KK, Wang J, Goodman JV, Larner SF. Antibody bound synthetic vesicle containing molecules for deliver to central and peripheral nervous system cells. US20110097392, 2011.
117. Viladot Petit JL, Delgado González R, Fernández Botello A. Lipid nanoparticle capsules. WO2011116963, 2011.