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Important Pharmaceutical Applications of Man-Made Lipid Nanocarriers for Sustained Drug Delivery and Future Outlook

Chukwuebuka Emmanuel Umeyor*; Emmanuel Maduabuchi Uronnachi

Nanomedicines and Drug Delivery Research Group, Department of Pharmaceutics and Pharmaceutical Technology, Nnamdi Azikiwe University, Awka 422001, Anambra State, Nigeria

Corresponding Author: Chukwuebuka Emmanuel Umeyor

Department of Pharmaceutics and Pharmaceutical Technology, Nnamdi Azikiwe University, Awka 422001, Anambra State, Nigeria.

Tel: +234-8063299850; Email: ec.umeyor@unizik.edu.ng

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Abstract

Recently, there has been increasing attention in the application of novel drug delivery systems (NDDS) based on lipids to enhance the Pharmacokinetic (PK) and Pharmacodynamic (PD) profiles of drugs, and tremendous successes have been reported due to the systematic application of these smart nanocarriers. These lipid-based nanocarriers promise excellent drug payload, improved bioavailability of entrapped drugs, and impart sustained release of these drugs. The design and development of these versatile nanocarriers is a novel area of research that offers new hope in drug delivery, especially those with very low clinical use. This chapter will attempt a discussion on the various trends in lipid-based drug delivery, as well as a careful survey of the different man-made lipid nanocarriers that have been applied for sustained drug delivery. Toxicity and ethical issues as well as future prospects in the design, development and applications of lipid nanocarriers for sustained drug delivery will be considered.

Keywords: Nanoparticles; Lipids; Sustained release; Nanocarriers; Drug Delivery.

Introduction

Since the famous lecture by Richard Feynmann in 1959 during which he declared the existence of “plenty room at the bottom”, researchers have explored the possibilities abounding in nanotechnology. These researches have yielded many positive results culminating in the use of nanoparticles for detection, mitigation, treatment and monitoring of diseases and disease progression. The size of these particles make them an interesting option for targeted delivery and reduced dosing of drugs, while a proper engineering of these particles would result in sustained release particles. The eutopic feeling generated by nanoparticle use is however not complete as researchers have to worry about the possible toxic effects arising from the use of inorganic particles (like metals) for diagnostic and therapeutic (theranostic) applications. Lipid particles thus become useful in this regard. Their biocompatibility and non-toxicity make them an interesting option for drug delivery.

Lipids are unique in their ability to be adapted to different routes of delivery: oral, dermal, transdermal, parenteral, making them very versatile. In addition, lipid formulations can be modified in various ways to meet a wide range of product requirements as per the disease condition, route of administration, cost, product stability, toxicity, and efficacy [1]. In delivering drugs orally lipids have gained much interest in their ability to improve absorption of poorly absorbed drugs (especially BCS class II drugs) [3]. Their biocompatibility and relative non-toxicity make them suitable for parenteral delivery, while their similarities with membrane lipids and a non-irritant, non-allergenic nature ensure that dermal delivery would pose little problems.

Man-made lipid-based particulate drug delivery systems (LPDDS)

Lipids have been explored in diverse ways for drug delivery and show remarkable properties in sustaining drug delivery as well as enhancing bioavailability at the nanoscale. Medically, nanolipids are important and unique due to their ability to absorb/adsorb other compounds as well as a higher surface to mass ratio than that of other colloidal particles. Depending on the route of administration, intended use, and method of production, lipid nanoparticles have been engineered to fulfill diverse needs and therefore are classified accordingly. They are formulated using mainly lipids and emulsifiers (Table 1). Some of the common lipid-based nanocarriers are discussed below.

Table 1: Some commonly used excipients in the production of lipid-based nanocarriers.

LIPIDS	
Class	**Examples**
Fatty acids	Oleic acid, palmitic acid, stearic acid, linoleic acid
Long chain triglycerides	Corn oil, soybean oil, safflower oil, olive oil, peanut oil, sesame oil.
Medium Chain Triglycerides (MCT)	Glyceryl tricaprylate/Caprate
Propylene glycol esters	Propylene glycol monocaprylate, propylene glycol monolaurate
Monoglycerides/Diglycerides	Glyceryl caprylate/Caprate, Glyceryl monocaprylate, Glyceryl monooleate.
Waxes	Beeswax, Carnauba wax
Hard fats	Witepsol H® 35, Witepsol W® 35, Witepsol E® 85, Witepsol S® 51, Witepsol S® 55 , Glyceryl monostearate (Imwitor® 900), Glyceryl behenate (Compritol 888 ATO), Glyceryl palmitostearate (Precirol® ATO 5)
Low HLB (<10) Emulsifiers	
Common names/type	**Examples**
Phosphatidylcholine (PC) and phosphatidylcholine solvent mixtures	Phosphatidylcholine (PC); PC in propylene glycol; PC in Medium Chain Triglycerides (MCT); PC in safflower oil/ethanol.
Unsaturated polyglycolized glycerides	Oleoyl macroglycerides; Linoleoyl macroglycerides
Sorbitan esters	Sorbitan monooleate, sorbitan monostearate, sorbitan monolaurate, sorbitan monopalmitate
High HLB (>10) Emulsifiers	
Common names/type	**Examples**
Polyoxyethylene sorbitan esters	Polysorbate -20; polysorbate -40; polysorbate- 60; polysorbate- 80
Polyoxyl castor oil derivatives	Polyoxy 35 castor oil, polyoxy 40 hydrogenated castor oil
Polyoxyethylene, polyoxypropylene block copolymer	Poloxamer 188, Poloxamer 407, poloxamer 182, Poloxamer, 237, Poloxamer 238, Poloxamer 338, Poloxamer 908
Saturated polyglycolized glycerides	Lauroyl macroglycerides, stearoyl macroglycerides
PEG-8 Caprylic/Capric glycerides	Caprylocaproxyl macrogolglycerides

Adapted with slight modifications from Cannon (2011) [2].

Solid lipid nanoparticles (SLN)

Speiser, otherwise known as the 'father' of nanoparticles, introduced the technology of formulation of nanoparticles in the early 80s. He actually produced microparticles with his co-workers when he applied high-speed stirring or shearing in the formulation of an o/w emulsion making use of a lipid melt and a hot aqueous surfactant solution. On cooling the obtained emulsion, solid particles recrystallized at room temperature, which when characterized, were in micrometer scale, giving Solid Lipid Microparticles (SLM). The production protocol adapted by Speiser and his research group was unable to produce particles in the submicron size [4,5]. The application of High Pressure Homogenization (HPH) in the early 90s revolutionized the manufacture of nanoparticles and significantly improved both the quality and size of SLM by producing more particle disruption and nanosizing, compared to high-speed shearing Speiser and his co-workers used. This process led to the production of Solid Lipid Nanoparticles (SLN). An interesting characteristic of SLN produced using HPH compared with high-speed shearing is the presence of low amount of microparticles [6,7]. This landmark research report by Speiser and co-workers opened new frontiers in formulation science, and many years after the report was made, SLN have continued to attract increasing attention among formulation scientists, especially due to the ease and efficacy of its production. Furthermore, it has been claimed that SLN possess all the advantages of other colloidal drug carriers but did not inherit their disadvantages. Some of these advantages include high drug entrapment capacity, increased drug stability, avoidance of organic solvents, etc. However, several

evidences abound of serious limitations inherent in the production of SLN, which have lowered its application in drug delivery. Identified limitations in the production and use of SLN include particle growth due to Ostwald ripening, high aqueous content of the dispersion, unpredictable gelation tendency, etc [8]. HPH is made up of two major techniques: hot and cold homogenization techniques, and these are used for the production of SLN by incorporating the drug into the bulk lipid by dissolving the drug in the lipid melt. Other methods of production of SLN include solvent emulsification/evaporation, solvent injection, dilution of microemulsions or liquid crystalline phases, etc. SLN also have the potential of sustained release of drugs, improved bioavailability of both hydrophilic and lipophilic drugs, and can be used for the delivery of drugs parenterally, orally, topically, rectally, etc. [9].

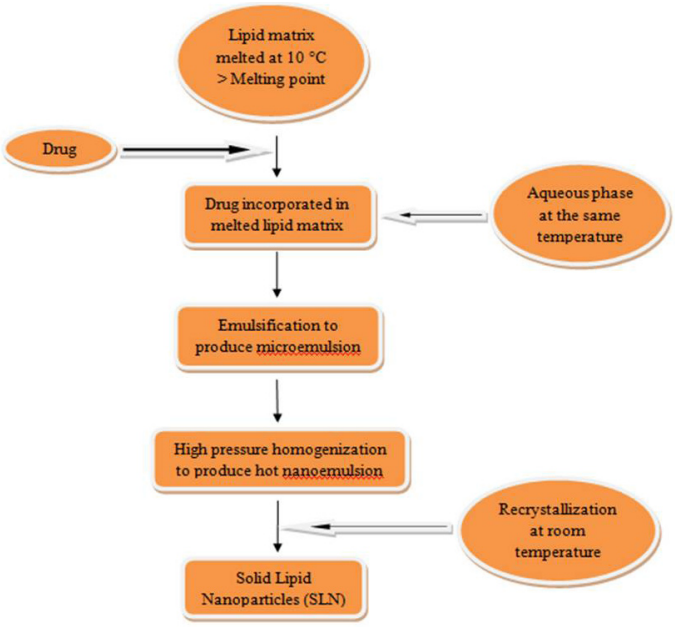

Figure 1: Production of Solid Lipid Nanoparticles (SLN) by high pressure homogenization technique.

Liposomes

Due to the urgent need to design drug carrier systems that will act as effective delivery systems and alter the biodistribution of drugs such that when administered, a therapeutic fraction reaches a target site, liposomes were introduced in the early 60s. Liposomes are particulate or colloidal carrier systems which spontaneously form when lipid bilayers are hydrated in aqueous media [10]. Liposomes are formulated with biocompatible and biodegradable lipid bilayers which enclose an aqueous core, and are separated from one another by aqueous domains (amphiphilic bilayers). This interesting structure of liposomes with alternating lipophilic and hydrophilic layers make it functionally possible for liposomes to encapsulate both lipophilic and hydrophilic drugs in the lipid layer, non-lipid layer or at the bilayer interface. Since its introduction, liposomes have been used to encapsulate vaccines, antibacterials, antimalarials, antifungals, nucleic acids, etc. [9,11].

Liposomes have been classified based on a number of factors. On the basis of the ability of liposomes to interact with cells, liposomes are classified into sterically stabilized or long-circulating or non-reactive liposomes and cationic or reactive liposomes. Considering their composition, liposomes are classified into: conventional, pH sensitive, cationic, immunoliposomes, and long-circulating liposomes. When the size and number of bilayers are considered, liposomes are classified into Multi-lamellar Vesicles (MLV), Large Unilamellar Vesicles (LUV), and Small Unilamellar Vesicles (SUV). In the use of liposomes for sustained drug delivery, some physicochemical parameters are considered. They include: bilayer fluidity, surface charge density, surface hydration, and liposome size. These parameters are very important because they influence in vivo behavior of liposomes and the release pattern of the entrapped drug [10].

Nanostructured lipid carriers (NLC)

Nanostructured lipid Carriers (NLC) is classified as a second generation lipid nanoparticles which were developed to alleviate the limitations affiliated to SLN, due to their highly ordered crystalline structure leading to the formation of low-energy βi and β lipid modifications which encourages low drug entrapment and drug expulsion [12]. As an improvement on SLN, NLC provides an advanced and convenient sustained release drug delivery system for various bioactives. In the preparation of NLC, lipid molecules with varying spatial orientation are blended together to create an imperfect matrix. Essentially, the production of NLC which involves the addition of a liquid lipid (oil) to a solid lipid leads to the creation of a less-ordered crystal lattice with numerous internal defects. Due to the many imperfections created in the lipid matrix core of NLC, limitations encountered in SLN such as low drug encapsulation and drug expulsion are resolved, as a high amount of guest drug molecules are readily accommodated [13,14]. In addition, NLC also have lower water content and less tendency of unpredictable gelation. They exert a controlled release effect and increase chemical stability of incorporated drugs. Furthermore, they are easily scalable [15]. NLC can be classified into I, II, and III types, on the basis of liquid lipid (oil) content and the ability of the lipid matrix to crystallize at room temperature. Class I NLC has lower liquid lipid (oil) content than the solid lipid component. Upon mixing, the structural differences in the lipids lead to the creation of highly disordered, imperfect matrix during crystallization desirable for high drug encapsulation. In class II NLC, the amount of oil is higher than the solid lipid. The oil component is used to prepare a nanoemulsion which is allowed to crystallize at room temperature before it is mixed with the solid lipid. This process leads to the formation of oily compartments which encapsulate drugs, but with increased solubility in the oily component of the matrix. For class III NLC, the lipid matrix is solid at room temperature but does not crystallize, and this helps to increase drug entrapment in the matrix [16-18].

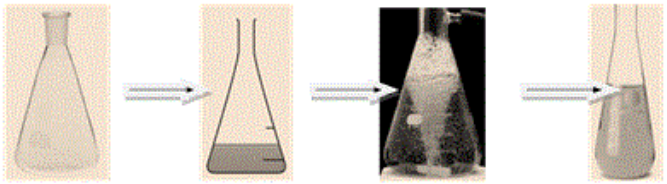

Figure 2: Production of Nanostructured Lipid Carrier (NLC).

Lipid nanoemulsions (LNE)

As formulation scientists were putting in a great deal of efforts to resolve the 'emulsion crisis' as to what defines the clear differences between a microemulsion and an emulsion, Lipid Nanoemulsions (LNE) was introduced as a drug delivery system with high potential for sustained drug delivery. As a delivery system which results from an interchange of solid lipids with liquid lipids in the presence of aqueous surfactants (and sometimes,

co-surfactants are added to increase solubilization capacity) under high shear mixing or mechanical extrusion, LNE is renowned as a competent pathfinder in the delivery of both lipophilic and hydrophilic bioactives leading to improved bioavailability due to increased solubility of these drugs in the delivery system [19,20]. LNE are oil-in-water (o/w) emulsions with mean particle diameters ranging from 50 to 500 nm, and the guest drug molecule is localized in the water or oil core of the emulsion. It is also referred to as submicron, ultrafine or mini- emulsion, formulated using biocompatible and biodegradable food-grade oils and GRAS (generally regarded as safe)-rated excipients [21].

Some researchers are of the view that there are no clear distinctions between nanoemulsions and microemulsions, since they are oil-in-water (o/w) dispersions, have small droplet diameter and narrow droplet size distribution, and are of low viscosity [19]. Conversely, there are certain differences between nanoemulsions and microemulsions. Nanoemulsions as structured liquid systems are produced following a deliberate application of microfluidic and ultrasonic techniques in emulsification leading to the rupturing of larger emulsion droplets to nanoscale droplets. Also, despite their metastable nature, nanoemulsions can maintain their physical state over a long period of time due to the stabilizing presence of surfactants and co-surfactants which act to inhibit emulsion droplets aggregation. In contrast to microemulsions, nanoemulsions are not lyotropic liquid crystalline phases, and do not form spontaneously because an external shear must be consciously applied so as to rupture microscale droplets into nanoscale ones [22]. Furthermore, LNE are produced by adapting the technologies used for the manufacture of SLN including High Pressure Homogenization (HPH), microfluidization, phase inversion temperature technique, solvent displacement/injection method, and self-nanoemulsification method. It is also used for the delivery of drugs through the oral, transdermal, pulmonary, nasal, and parenteral routes, etc. as well as in vaccine and gene delivery [21].

Cochleate nanocarriers

Cochleates, as nanoparticulate drug delivery system, were first studied and reported by Dimitrious Papahadjopoulos and his research team in the early 70s. In their report, they described cochleates as precipitates formed when calcium reacts with negatively charged Phosphatidylserine (PS). These precipitates were named 'cochleates' meaning 'shell' in Greek, due to their rolled-up structure [23]. Cochleate drug delivery system was introduced to provide answers to some oral drug delivery challenges such as poor bioavailability of lipophilic drugs, and enzymatic degradation of drugs in the Gastrointestinal Tract (GIT). Since its introduction, several researchers have reported the manufacture of cochleates using single negatively charged phospholipids or their admixtures including Phosphatidic Acid (PA), Phosphatidylcholine (PC), and phosphatidylethanolamine. Other phospholipid derivatives such as galactosphingolipid hydroxyl fatty acid cerebroside have been reported to form cochleate by thermal mechanical treatment of glycol suspensions [24]. In addition, cochleates are also easily formed from Small Unilamellar Vesicles (SUV). However, Multilamellar Vesicles (MLV) can also lead to the formation of cochleate through destabilization of the outer layer of PS by calcium ion easily with higher access of the ions into the inner PS bilayers [23]. Several technologies have been explored for the production of cochleate nanoparticles including the hydrogel method based on aqueous-aqueous emulsion system, and this yields small nanosized particles. Trapping method, which is very useful for the entrapment of hydrophilic and lipophilic drugs, has been described and it's characterized by the aggregation of particles. Cochleates are also produced using solvent drop method designed mainly for encapsulating lipophilic drugs using solvents such as Dimethylsulphoxide (DMSO) and Dimethylformamide (DMF) [25].

Lipid nanocapsules (LNC)

Polymeric nanocarriers have continued to elicit a high level of interest owing to their potential use as drug carriers and their ability to control the release of encapsulated drugs [26]. Essentially, the class of polymeric nanoparticles that have assumed this popularity is Lipid Nanocapsules (LNC). These are regarded as vesicular carriers consisting of an oily core surrounded by a thin polymeric wall [27]. LNC represents a very important class of polymeric nanocarriers capable of effectively entrapping and delivering therapeutic amount of a variety of drugs, proteins and peptides, while increasing their stability and decreasing their toxicity. Several researchers have developed LNC using a dispersion of sorbitan monostearate, medium chain triglyceride e.g. capric or caprylic acid as core, enveloped by polymers such as poly (Ɛ-caprolactone), an aliphatic polyester [28,29]. Properties of LNC include increasing the solubility of poorly water soluble drugs, targeting the encapsulated drugs to specific sites and prolonging the blood levels of drugs. LNC are prepared either by *in situ* polymerization, including polymerization in emulsion and interfacial polymerization, or by polymer self-assembly, such as nanoprecipitation and interfacial deposition of the polymer, salting-out and emulsification-diffusion. In summary, these methods involve the use of an organic and an aqueous phase to structure the colloidal dispersion [29]. LNCs have been used systemically to treat acute and chronic inflammation, multiform glioblastoma, neuroinflammation, in the food industry, as well as for topical drug delivery [30-32].

Challenges in the use of lipid-based nanocarriers as drug delivery systems

Despite their numerous advantages in drug delivery, some challenges are met in the use of lipid nano-carriers as drug delivery agents. These challenges include:

a) Possible clearance by the reticuloendothelial system (RES)

Owing to the size of the nanoparticles, oftentimes, they are identified as "foreign bodies" by the cells of the RES thereby leading to a rapid clearance from the systemic circulation. This leads to a short circulation time for the nanoparticles. A possible method for reducing this clearance is by linking the nanoparticles with polyethylene glycols (PEGylation). PEGylation has been carried out by several researchers as a means of increasing the circulation time of these nanoparticles by evading the RES. PEGylation confers hydrophilicity on the nanoparticles. It also prevents immunoprotein adsorption and phagocytosis by macrophages [33]. In addition to PEGylation, limiting the particle sizes of the nanoparticles to 200 nm or less can also enable them evade the RES, due to the fact that these cells do not recognize particles lower than 200 nm as "foreign" [9].

b) Polymorphic transitions during storage

Due to the nature of the starting materials (lipids) for these nanocarriers, there is a great tendency for polymorphic transitions to occur during storage. Different polymorphic forms of triglycerides exist: α, γ, β, and β^1 forms. Polymorphic transitions

usually occur from the metastable α form via the γ and β^1 intermediates to the stable β form. This β form is usually less amorphous and more crystalline than the metastable forms. The consequence of this more crystalline form is an ordered matrix with less space for drug entrapment leading to expulsion of some of the entrapped drug. Another effect of these modifications is a loss in the ability of the vesicles to sustain the release of the entrapped drug [9]. Polymorphic changes can be affected by factors like temperature and solvent effects.

c) Gelation tendency

The tendency to gel can be induced by morphological changes and transitions from spherical to platelet like forms [9,34]. The emulsifier, lipid type, and concentration of both emulsifier and lipids could lead to gelation of nanoparticles. In addition, processing parameters like homogenization speed and time could induce gelation in nanoparticles. A postulate for gelation is the stacking of the lipid nanoparticle platelets leading to self-association [9,35].

d) Supercooling of nanoparticles

This could occur with the use of lipids that are solids at room temperature e.g. triglycerides. During nanoparticle formation, especially via the homogenization method, super-cooled melts could occur [9].

e) Physical stability

Nanoparticles due to their size, characteristically exhibit Brownian motion. Differences in particle size and aggregation of particles could also lead to a phenomenon known as "Ostwald ripening". This is particularly visible in nano-dispersions. Such instabilities could impact negatively on the shelf-life of the nanoparticles. Surface stabilization can be achieved with the use of emulsifiers in suitable amounts [9].

f) Large aqueous volumes of nanodispersions

Typically, the aqueous portions of nanodispersions contain water contents > 70 %. This large volume predisposes them to instabilities. Several methods have been explored in removing the water. These include lyophilization and spray-drying. For an effective lyophilization process, suitable cryoprotectants and loprotectants are included in the formulation. These would preserve particle integrity during the lyophilization process [9]. Spray drying is also an efficient process of transforming nanodispersions into powders [36].

g) Existence of more than one colloidal structure in the system

Similarities in method of production of different nanocarriers as well as the use of similar excipients have led to the possibility of more than one type of nanocarrier existing in a system e.g. presence of liposomes in NLCs. The formation of additional colloidal systems may arise from a redistribution of emulsifiers from the particle into the aqueous phase thus giving rise to the self-assembly of liposomes and other colloidal structures. Formation of drug nanocrystals could occur if the saturation solubility of the lipid for the drug is exceeded [9].

h) Sterilization

Since some of the lipid nanocarriers formulated is intended for parenteral administration, sterilization of the preparations before administration becomes pertinent. Among the numerous sterilization methods available, filtration using membrane filters (usually, a pore size of 0.22 μm) is used commonly. This sterilization process preserves the structural integrity of the lipid nanocarriers and has been employed by several researchers. A limitation of the filtration method is the size of the nanoparticles, as those possessing sizes closer to or greater than the membrane filter's pore size (0.2 μm) would clog the filter [37]. In recent times however, moist heat has been successfully utilized in sterilizing nanoparticles without affecting the integrity of the vesicles [38].

Characterization of lipid nanocarriers

Determination of the characteristics of lipid nanocarriers is essential to ensure the formulation of nanodispersions for the right application, and with the desired properties. At the commencement of lipid nanocarriers characterization, the major concern is the stability profile of the nanodispersions, especially considering the propensity of lipids to undergo several polymorphic transitions and particle size variation at the nanometer scale. Since lipid nanocarriers are complex systems with several particles of different sizes and distributions coexisting in the nanodispersion coupled with the challenge of polymorphic transformations, several techniques utilizing sophisticated equipment have been drafted for detailed characterization of lipid nanocarriers so as to give acceptable impression of their orientations, highlights and associated challenges.

Measurement of particle size

Particle size determination is an important characterization technique for lipid nanocarriers as it will help to confirm that dispersions in the colloidal size range were obtained during the formulation process, as well as ensure that the size range of the particles were retained upon further processing such as during lyophilization, sterilization, etc. and storage. In addition, particle sizing is important in determining the extent of interaction of nanoparticles with biological tissues and the effect of such linkage. It is also essential in formulation development and optimization. Determination of particle size of lipid nanocarriers can be accomplished using nanoscale microscopic methods, but light scattering methods have assumed prominence in this procedure e.g. Photon Correlation Spectroscopy (PCS) or Dynamic Light Scattering (DLS) which analyzes flip-flops in the intensity of light scattering by the test specimens due to Brownian motion [39], giving average size of diameter of the nanoparticles. Particle size can also be measured using volume laser diffraction, a volume-based technique, which analyzes particle sizes by determining volume of equivalent spheres and weighted mean of volume distribution of the particles [21]. It is essential to note that lipid nanocarriers should be diluted appropriately before size determination. Although this may be acceptable practically, it should be known that particle redistribution due to dilution may alter the original orientation of the dispersion thereby rendering the characterization procedures subjective [40,41].

Determination of surface morphology

Determination of surface morphology of lipid nanocarriers is a very important step in defining particulate orientation and structure of nanodispersions. Of special interest is the fact that particulate shape affects drug entrapment and subsequent release profile in lipid nanocarriers because spherical nanoparticles have been found to be easily stabilized with minimum concentration of surfactants due to its small specific surface area, and offers the potential for controlled release of drugs [42]. Morphology of lipid nanocarriers can be studied using Scanning

Electron Microscopy (SEM), Transmission Electron Microscopy (TEM) or cryo-TEM, optical microscopy, Atomic Force Microscopy (AFM), etc. SEM finds greater application in the analysis of shape and surface orientation of Solid Lipid Microparticles (SLM), as it is currently scarcely used for lipid nanodispersions. Through point by point scanning of the test specimen, a three-dimensional impression of the structural features of the sample is obtained and analyzed using image analysis software [43-45]. TEM on the other hand, provides direct image of the structural architecture of lipid nanoparticles. TEM provides valuable structural information that complements data from other analytical techniques giving a complete picture of the particle size, shape or the presence of any other colloidal structures present in lipid nanocarriers. In addition, it provides images with higher resolution, and analysis could be done using image analyzers, giving vital information on the sizes and size distribution of lipid nanodispersions [21,46]. In order to resolve the presence of complex colloidal structures in lipid nanocarriers, cryo-electron microscopy or cryo-TEM is applied where a cryogen e.g. liquid propane or nitrogen is used to enable the observation of specimens in their original nature [47,34]. Optical microscopy can also be used to analyze lipid nanocarriers sparingly due to the sub-micron particle sizes. However, it can be used for the detection of phase-separated crystalline biomolecules and for the characterization of microparticulate contaminations. In addition to this, AFM provides useful information about physicochemical properties of excipients used in the formulation of lipid nanocarriers, which influence the *in vivo* performance of the drug carrier system [48].

Measurement of zeta potential

To measure the zeta potential of nanocarriers involves the determination of the net attractive or repulsive forces existing between particles in lipid nanodispersion. Zeta potential is a physical property exhibited by particles of a colloidal dispersion. It can be used to optimize lipid nanoformulations as a good knowledge of the zeta potential of dispersion can reduce or eliminate time and materials wastage in trial formulations. It is determined using an instrument called zetasizer, which provides information regarding the stability profile of lipid nanocarriers against aggregation because it gives a very good index of the magnitude of interaction between colloidal systems. Zeta potential of a lipid nanocarrier also affects the *in vivo* behavior of an encapsulated drug due to its interaction with systemic electrolytes of biological fluids [49]. In a weak electric field, the zeta potential can be derived from the Helmholtz-Smoluchowski relation:

$$\mu = \varepsilon\zeta/\eta$$

Where μ is the electrophoretic mobility, ε is the permittivity, ζ is the zeta potential, and η is the viscosity of the dispersion medium [42]. When stability of surface modified colloidal systems e.g. pegylated nanocarriers is considered from the measurement of zeta potential, it is essential to take into account that surface charge may not be the only stabilizing mechanism as steric effect has to be considered. Investigation of such systems reveals the presence of a range of zeta potential values. However, these set of data are comparative from one to another, and results in highly stable lipid nanodispersion because the PEG moiety interferes with the electrophoretic mobility of the particles of the colloidal system [49,50].

Study of polymorphism and crystallinity

Since nanocarrier drug loading capacity and modified release characteristics are influenced by polymorphism and crystallization, studying these factors is of great importance in the characterization of lipid nanocarriers, especially when new drugs are incorporated or novel methods are tested [9,36]. This is because increasing the order of crystallinity of the lipid nanoparticles limits drug loading capacity and vice versa. The organization of alkane chains present in lipids into different packing patterns is largely implicated for the occurrence of polymorphism and crystallization in lipid nanocarriers. As a result, hexagonal (α), orthorhombic (β'), and triclinic (β) structures are obtained [51,52]. Lipid crystallinity increases from α to β and β', and increasing order of crystallinity reduces drug entrapment as seen in the more stable β and β' forms of lipids, but this is usually high for less ordered polymorphic forms of lipids e.g. metastable α-forms of lipids [53]. This is due to the reduction in the degree of disorderliness and the number of imperfections created in the lipid core, as a result of the formation of a more perfect lattice and surface-deposition of drug molecules leading to drug expulsion. Over the past decades, research reports have indicated that chain length of triglycerides used in nanocarriers production affect crystalline and polymorphic orientation of the nanoparticles. The use of long-chain triglycerides has being favored in the production of lipid nanoparticles than short-chain triglycerides, since unfavorable lipid transformation from α to β polymorphs occur slowly in the former than the latter. This suggests that for a lipid nanodispersion to be defined as efficient and optimal means that there should be the presence of a high percentage of α crystals and a low concentration of β/β' polymorphs [54]. Furthermore, it has been shown that the addition of surfactants in the hot lipid nanodispersion affect the time course of lipid nanocarriers undergoing polymorphism after crystallization [36]. In a study, it was observed that surfactants influence crystallization temperature of lipid nanoparticles, leading to homogenous or surface heterogeneous nucleation. However, this submission depends on the lipophilicity or hydrophilicity and the presence or absence of charges on the surfactant used in the nanoparticulate formulation process. Based on this, it has been suggested that increased lipophilicity of lipid nanocarriers increases the extent and rate of nanoparticle crystallization, and vice versa. Similarly, production of lipid nanocarriers with non-ionic surfactants increases the rate of particulate crystallization, while this occurs slowly with ionic surfactants [55]. From the above, it is reasonable to infer that, for optimal nanoparticle production, a formulation scientist needs a good understanding of the behaviour and transition mechanism of starting lipids and the type of surfactant to be used.

Crystallization and polymorphism in lipid nanoparticles are widely studied using Differential Scanning Calorimetry (DSC) and X-Ray Diffraction (XRD). XRD is used to characterize the polymorphic structure present in lipid nanocarriers. It provides information about the length of long (wide-angle XRD) and short (small-angle XRD) spacing in the lipid core at a given temperature, allowing crystalline and amorphous systems to be determined. DSC, on the other hand, provides information about the crystallinity and physical state of lipid nanoparticles. Analysis of DSC thermograms helps in the determination of the fusion enthalpy and temperature of the nanoparticles, and it is established that a high fusion enthalpy indicates an ordered system (perfect lattice) which requires high amount of energy

to overcome the cohesive forces in the crystal lattice. For objectivity in assigning accurate polymorphic data to lipid nanoparticles, DSC data should be supported by information obtained from XRD analysis [9,56-58].

Analysis of drug release and stability of lipid nanocarriers

Available data have shown that lipid nanocarriers exhibit prolonged *in vitro* release, and subsequent controlled *in vivo* behavior for various biomolecules. Major factors which influence drug release from lipid nanoparticles are physicochemical properties of the drug itself, particulate size, composition of lipid core, distribution of drug molecules throughout the lipid core, type and concentration of surfactants used, production technology adapted and process parameters [59]. Generally, information on release studies of lipid nanoparticles have been very few due to difficulties experienced by formulators in conduction such experiments. Most times, *in vitro* release studies involve processes such as filtration and centrifugation of colloidal dispersions of nanoparticles, or dialysis, all of which serve as obstacles in release studies [60]. Burst effect has been identified as a serious challenge experienced in drug release studies from nanocarriers due to drug accumulation on the surface of the particles. Although it can be considered to be useful in the sense that it helps in the release and delivery of an initial dose of drug, it is often seen as a failure of a presumed controlled drug release process leading to incomplete drug release [61,62]. In contrast to this, entrapment of drug in the lipid core encourages prolonged release of drug. It has been suggested that burst release of drugs from nanoparticles can be controlled by controlling drug solubility in the aqueous phase during production which, in turn, can be controlled by regulating the temperature of production as well as excipient concentration e.g. surfactant, used in the formulation. Nanoparticulate production at high temperature and surfactant concentration increases the establishment of burst effect in nanoparticles, whereas nanocarriers produced at regulated conditions of temperature and surfactant avoids drug partitioning into aqueous phase and subsequent re-partitioning into the lipid phase, showing no burst release of drugs [54]. In an experiment to study drug release patterns of tetracaine and etomidate from SLN, burst release of both drugs within a few minutes was observed for both liquid and freeze-dried SLN. It was suggested that the fast release of the drugs from the SLN could be due to large surface area of particles, a high diffusion coefficient due to small molecular size, low viscosity in the matrix and a short diffusion distance covered by the drugs [63].

Lipid nanoparticles provide great opportunities in the delivery of drugs to improve their bioavailability and therapeutic efficacy. The loading of drug molecules in lipid nanocarriers depends on the ability of the carrier system to preserve and protect its integrity and that of the encapsulated drug. Therefore, characterization steps involved in the formulation of lipid nanocarriers should include information regarding the stability of the loaded drug to ensure its safety and efficacy. This is necessary because some of the production techniques of lipid nanoparticles such as production at high temperatures, mechanical agitation, high pressure homogenization, use of organic solvents, may damage the structure of an encapsulated drug with attendant losses in the physiological activity of the drug. Stability of nanocarriers should be considered during packaging, storage (due to the effects of sedimentation, aggregation, and crystal growth) or during shipping. This is because factors such as particle aggregation may lead to embolism as a result of the blockage of blood capillaries and obstruction of blood flow in parenteral drug delivery. Stability of lipid nanoparticles is mainly affected by processes which could be chemical or physical, though protection of an encapsulated drug from harsh biological environment is equally considered important as biological degradation due to the route of administration of the particles may negatively affect the stability of the nanocarrier. Physical instability in lipid nanocarriers is mostly due to lipid modifications or modifications in the colloidal dispersion. A minor modification of the lipid applied in the production of nanocarriers may change the structure of the resulting nanoparticles, and this may change the drug-loading capacity, surface or interfacial, and drug release properties [64].

Lipid modifications may be due to failure of some lipid materials used in the production of nanocarriers to crystallize in the colloidally dispersed state as well as the transformation of lipid particles from the less stable α-form to more stable β-form (crystallization-polymorphic transitions). However, these changes could be explained through thermoanalytical characterization of the lipid nanocarrier to reveal the degree of crystallinity of melt-homogenized nanodispersion, crystallization behavior, time course of polymorphic transitions, enthalpy and fusion temperature using combined data obtained from Differential Scanning Calorimeter (DSC) and X-Ray Diffractometer (XRD) [54,65,66]. Gelling causes rapid modifications in lipid nanocarriers due to increase in kinetic energy of the particles leading to particle collision, and it is irreversible. This phenomenon could be caused by production of nanocarriers using a high concentration of lipid, high ionic strength materials, exposure of the nanoparticles to high temperature, light and mechanical agitation. However, gel formation in lipid nanoparticles could be attenuated by the addition of co-surfactants or co-solvents or by the production of lipid nanoparticles at optimum process parameters determined from carefully designed pre-formulation studies [67,68]. Physical instability due to colloidal modifications results from Ostwald ripening caused by dissolution and deposition of smaller particle crystals on larger particles leading to particle growth, and coalescence [69].

Chemical instability in lipid nanocarriers is of great importance in the nanoparticulate characterization as it affects the integrity, safety and efficacy of the carrier and encapsulated drug molecules. Good knowledge of the stability properties of a drug molecule as well as the nanocarrier system would help in proper selection of appropriate process parameters, formulation techniques, packaging and storage conditions, which are also essential for regulation [70]. Triglycerides undergo hydrolytic reactions to produce mono- and di-glycerides with free fatty acids. The first report on chemical stability of lipid nanocarriers involves the use of the triglycerides, Dynasan 112-188 to produce SLN and comparing with SLN made from a blend of mono-, di- and triglycerides, Softisan/Imwitor and Witepsol S51/Witepsol S55 followed by evaluation of their chemical stability using Gas Chromatography (GC). GC revealed that SLN formulated with triglycerides were more stable than those obtained with mono- and di-glycerides, due to the ability of the triglycerides to be localized in the lipid core [54]. According to the FDA, stability assessment of drug carrier systems could be carried out by means of a validated quantitative analytical procedure called Stability Indicating Method (SIM), which indicates how the stability of drug substances and their carrier systems changes with time. SIM is designed to evaluate the rate and extent of decrease in the amount of drug molecules in drug carrier systems due to degradation without interference from degra-

dation products, impurities and excipients. An HPLC equipped with a UV detector is widely used in SIM studies and it avails important information to assist pre-formulation studies, stability studies, and the development and adoption of proper storage requirements [71,72].

Toxicologic aspects of lipid-based nanocarriers

The introduction and development of lipid nanocarriers revolutionized and provided great opportunities in drug delivery to improve the pharmacokinetic and bioavailability profiles of bioactives. The exponential rise in the biomedical applications of lipid nanocarriers for therapeutic and diagnostic (theranostic) purposes have highlighted the importance of considering the potentially unpredictable and adverse outcomes in the use of lipid nanoparticles by humans. This is especially vital considering that due to their small size, lipid nanocarriers have a large surface area to volume ratio which makes it possible for them to interact with many biological processes and tissues.

Toxicity of nanocarriers refers to the propensity of the particles to affect the normal physiological processes, and directly interrupt the integrity and functions of the biological system adversely. Nanoparticulate toxicity is usually due to the active drug encapsulated and in some cases, toxicity effects can be linked directly to the carrier system. Any drug delivery strategy that is biomedically applied to produce a therapeutic effect is also capable of producing undesirable, and sometimes, fatal adverse effects. The narrower the drug therapeutic index, the higher the risk of toxicity, and this risk increased when the formulation is non-specific in its actions [73,74].

Generally, toxicity of lipid nanocarriers could be related to physicochemical factors such as particle size, shape, surface charge, chemistry, composition, and stability effects; mechanistics such as oxidative stress, pro-inflammatory gene activation; and particle related factors such as administered dose, route of administration, and extent of tissue distribution [73]. Despite the fact that lipid nanocarriers are well tolerated by living tissues, since they are made from physiological and biodegradable lipid materials, some carrier systems formulated with certain lipid materials still pose lethal toxicity threats *in vivo*. Cationic lipid-based nanocarriers is one of such lipid-based systems associated with significant toxicity issues due to the structural orientation of cationic lipid molecules [75,76]. Typically, cationic lipids have a cationic head, a hydrophobic backbone and a linker region e.g. Dimethyldioctadecyl Ammonium Bromide (DDAB), N-[1-(2, 3-dioleoyloxy) propyl]-N,N,N-trimethyl ammonium chloride (DOTAP). Due to their orientation, cationic lipids can disrupt or damage the integrity of a cell membrane and irritate the exposed cell, cause vacuolization of cytoplasm, reduce the number of mitoses, cause cell shrinkage, lysis and sometimes, cell necrosis occur [77,78].

Interestingly, most of the toxicity studies on lipid nanocarriers involve the use of *in vitro* experimental designs in which dose-response effects produced by nanoparticles are measured and recorded, and sometimes, these *in vitro* results are extrapolated to represent certain *in vivo* effects. However, there are a few toxicological reports of lipid nanocarriers in animal models, which are the preferred systems for toxicologic evaluations, since *in vitro* cultures cannot replicate the complexity of an *in vivo* system or provide meaningful data about the response of physiologic tissues and organs to lipid nanoparticles [73,79], e.g. in a study, multiple high dose bolus injections of SLN equivalent to a 6-fold bolus injection of 100 g of SLN was administered to mice, and the histology of the hepatic and splenic tissues showed that the particles were well tolerated, with minor reversible alterations [80]. Similarly, in another study, NLC-based topical gel of aceclofenac was administered on the skin of wistar rats, and the gel did not produce any untoward effects on the skin of the rats [81]. Other reports have shown that lipid nanocarriers are biocompatible for improved drug delivery.

Pharmaceutical applications of lipid nanocarriers for sustained drug delivery

Lipid-based nanocarriers have been utilized since their introduction, for the sustained delivery of lipophilic and hydrophilic biomolecules so as to achieve different therapeutic benefits. Important pharmaceutical applications of lipid nanocarriers include:

a) Cosmetics and dermatologics delivery

Lipid nanocarriers are novel colloidal drug delivery systems with excellent and exciting cosmetic and dermatologic properties including enhancement of chemical stability of entrapped biomolecule, excellent skin hydration, enhanced skin bioavailability, controlled adhesion and occlusion, drug targeting, film formation on skin, whitening effects, absorption-enhancing effects, improved skin penetration, and controlled release properties [82,83]. Skin aging due to excessive exposure of the skin to Ultraviolet (UV) light is enhanced by certain environmental factors such as sun, heat, pollution and smoking [84]. However, lipid nanocarriers aid in reviving the tensile strength and softness of the skin by forming a protective barrier on the skin, making it water-resistant, reduce the effect of transdermal water loss and generally protect the skin against dehydration. These processes lead to the filling up of microscopic pores and indentations of the skin leading to skin smoothening, softening, emollience and removal of wrinkles. Therefore, lipid nanocarriers have been successfully applied for the sustained delivery of molecular sunscreens, UV blockers, anti-oxidants, analgesics, antimicrobial agents, etc [14,85-90].

It is proposed that lipid nanocarriers fluidizes the bilayer lipids of the stratum corneum, and facilitate the efficacious transport of active molecules through the stratum corneum into the deeper layers of the skin than conventional cosmetic products. For instance, the chemical stability of labile compounds like α-lipoic acid, α-tocopherol and β-carotene was enhanced when incorporated into lipid nanoparticles, while tretinoin became less irritating after entrapment in a lipid nanocarrier system [91-93]. In a study, α-lipoic acid encapsulated in lipid nanocarriers (NLC and SLN) demonstrated similar anti-oxidant property to the pure biomolecule with good physical and cytotoxic profiles. Similarly, the anti-aging property of curcuminoids was improved when compared with a conventional cream with no irritation [92,94]. These examples highlight the success of lipid nanocarriers in tackling skin aging, and their interesting potential as smart alternatives for the sustained delivery of cosmetics and dermatologics.

b) Ocular drug delivery

Drug delivery to the eye using lipid nanocarriers is a very important and challenging area of research in formulation science because the eye is the visual centre of the body and boasts of a robust immunological design. Drug delivery to the eye through the use of solution-type eye drops have experienced tremendous challenges and poor bioavailability with respect to the poor access of the posterior segment of the eye, and in treat-

ing vision-impairing conditions of the eye. Poor bioavailability of drugs from these conventional eye drops is mainly attributed to the pre-corneal loss factors which include tear dynamics, non-productive absorption, transient residence time in the cul-de-sac, and relative impermeability of the corneal epithelial membrane [8]. Consequently, for improved drug pharmacokinetics and targeting, these primary barriers in the eye need to be overcome. Also, drug redistribution through the vascularized regions of the eye needs to be controlled. Lipid nanoparticles have been shown to improve ocular bioavailability of drugs due to the propensity of these biocompatible carrier systems to produce excellent interaction with ocular mucosa, thereby prolonging the corneal residence time and increased absorption of the encapsulated drug. This property has also enabled drug targeting to the ocular centre, especially when the nanoparticles are surface-modified e.g. tobramycin, cyclosporine A, timolol, and pilocarpine [95,96]. In a study, diclofenac sodium was loaded in SLN and its release property was evaluated using human cornea construct. The researchers reported sustained release and improved permeation of the entrapped drug [97]. It has been shown that positively charged nanoparticles can be used to improve ocular drug delivery through corneal bioadhesion and drug permeation due to phagocytosis by corneal epithelial cells [98]. Similarly, *in vitro* permeation study of some poorly water soluble drugs loaded in SLN showed prolonged drug release [99].

c) Gene and vaccine delivery

Lipid nanocarriers have been shown to provide a suitable alternative for the sustained delivery of genes and vaccines. Besides sustained delivery, protection and facilitated transport of genes and vaccines, nanoparticles also have potential benefits of targeted delivery across membranes docking at tissues and modulate the release of their payload. They also ensure more effective recognition of antigens by immune cells [95,96]. Extensive reports have been made on the delivery of genes and vaccines using biodegradable polymeric nanoparticles and microparticles; however, polymeric nanoparticles present challenges of costs and the toxic organic solvents used for their production, which in turn, could lead to the deterioration of the integrity and structures of these biomolecules [100]. PLA/PLGA are the most commonly used polymers for the production of nanoparticles for the delivery of antigens and vaccines with the attendant advantages of macrophage recognition, prolonged immunity and sustained release. However, lipid-based nanocarriers have been shown to be better immunological systems providing both cellular and humoral immunity for both bacterial and viral antigens. Sometimes, protein-based antigens may be covalently-linked to triacylglycerols producing lapidated antigens which could mimic viral particles and self-assemble in water and enhance both humoral and cellular immune responses at systemic and mucosal levels [59,100]. Recently, lipid nanoemulsions have been researched upon for the delivery of Human Immunodeficiency Virus (HIV) vaccine through the nasal route in the ongoing fight against HIV [21]. This ground-breaking research clearly points to the fact that lipid-based nanoformulations could be the future vehicle for vaccine delivery in the fight against deadly viral infections e.g. ebola, zika, etc. due to their immunoadjuvant properties, especially when administered through the parenteral or mucosal routes for the delivery of vaccines.

d) Pulmonary delivery of drugs

The use of lipid-based nanoparticles for drug delivery to the lungs is still a virgin area for research, as pulmonary drug delivery has not been fully exploited owing to the sparse publications in this area. As a result, extensive study in lipid drug delivery through the pulmonary route is currently ongoing in various laboratories with a view to making this route of drug administration a popular alternative for non-invasive systemic delivery of biomolecules. In addition to its non-invasive nature, pulmonary drug delivery is interesting due to the large surface area (~ 100 m^2) of the lungs available for drug absorption, docking of administered drugs within the pulmonary tissue, circumvention of first-pass metabolism, the low thickness of the epithelial barrier, easy access to an extensive vasculature, and a relatively low enzymatic activity in the alveolar space compared with the Gastrointestinal Tract (GIT)/liver [73,101].

Lipid-based nanocarriers possess excellent physicochemical characteristics which make them suitable and appropriate delivery systems for the pulmonary route due to the correlation between their nanometer range diameter, biocompatibility, and deep-lung deposition ability. This encourages prolonged drug serum concentration and lung retention time. For the treatment of lung infections, lipid carrier systems have been studied for the delivery of budesonide, curcumin, amikacin, amphotericin B, ciprofloxacin, tacrolimus, itraconazole and beclomethasone [102-111]. Phospholipid-based SLN has been extensively studied for the delivery of drugs to the pulmonary tissue. This is due to the abundance of phospholipids in the deep recesses of the lungs and its essential role in breathing. Phospholipid-based SLN has been formulated and evaluated as a potential drug carrier system to the pulmonary area, and the system did not lead to production of pro-inflammatory mediators upon administration in mice. Similarly, quercetin-loaded Solid Lipid Microparticles (SLM) were formulated, characterized and evaluated for potential application in the treatment of asthma. Characterization studies showed that the microparticles fall within acceptable Mean Mass Aerodynamic Diameter (MMAD) values, the particles were stable after nebulization, and were mainly deposited in deep areas of the lungs [112]. Very recently, NLC has been studied as a potential delivery system for bioactives through the airways. NLC-loaded itraconazole were formulated for pulmonary delivery and found to have high entrapment efficiency with no particle size growth for formulations stored in both refrigerator and at room temperature. Nebulizing the itraconazole-loaded NLC from a jet stream had no influence on the particle size or the entrapment efficiency of itraconazole in the particle matrix [113]. The above examples show that lipid nanocarriers provide attractive basis for drug delivery due to the possibility of particle retention in the airways offering prolonged release and improved drug protection as well as better bioavailability in comparison to conventional dosage forms for pulmonary delivery.

e) Oral drug delivery

Orally administered drugs are the most commonly found in clinics. Drug administration through the oral route is available for all age brackets; however, it is commonly available for children and paediatrics. Lipid-based nanocarriers have been extensively researched for oral delivery of both lipophilic and hydrophilic drugs; however, these carrier systems are mainly applied for improving the poor solubility and bioavailability experienced by lipophilic drugs in the gastrointestinal milieu. This stems from the knowledge that intake of a fatty meal leads to prolongation of GIT residence time, stimulation of biliary and pancreatic secretions, stimulation of lymphatic transport, en-

hancement of intestinal wall permeability, reduction of metabolism and efflux activity, and alteration in mesenteric and liver blood flow; therefore, there could be a significant improvement in the oral bioavailability of drugs administered using lipid nanocarriers [12]. These nanocarriers are mainly formulated using triglycerides and upon administration, they are digested by pancreatic lipases into solubilized phases as mono- and di- glycerides. Absorption of these drug-laden lipids occurs through the transcellular or paracellular routes. The transcellular route of absorption involving the M cells in the Peyer's patches or enterocytes is the major pathway through the formation of chylomicrons into the lymphatic system. Size influences drug uptake in the lymphatic system, and smaller size particles favours higher absorption. Drug absorption using lipid nanocarriers is also improved due to the high adhesivity of the drug-bearing lipid particles to the gut wall; however, absorption through the M cells is greater [9,12,114]. Lymphatic absorption of nanoparticles depends on the length of the fatty acid chains, and reports have shown that Long Chain Triglycerides (LCT) of C-14 to C-18 promote lymphatic absorption than Medium Chain Triglycerides (MCT) [12]. Several hydrophilic and lipophilic drugs have been formulated as lipid-based nanocarriers to improve their solubility, absorption and bioavailability, or bypass the first-pass metabolic activities of the liver e.g. lovastatin, vinpocetine, cyclosporine, all-trans retinoic acid, curcumin, fenofibrate, insulin, etc [13,115,116].

f) Parenteral drug delivery

Lipid-based nanocarriers have been applied in the delivery of drugs ranging from subcutaneous, intraarticular, intramuscular to intravenous routes. Parenteral drug administration is favored in lipid nanocarriers due to their small size which is in the nanometer scale, and this is very important considering that these nanoparticles are syringed before administration. Consequently, the concern on the possibility of the occurrence of thromboembolism is adequately taken care of [8,9]. Lipid nanocarriers have been tactically engineered by surface modification using materials such as Polyethylene Glycol (PEG), poloxamines, poloxamers, etc. giving stealth characteristics for targeting drugs to intrinsic tissues such as spleen, brain, liver, lungs, and even diseased or cancer cells or tumors, where the nanocarrier systems docks and delivers its payload for prolonged blood circulation, resulting in controlled and sustained release effect, sometimes even post-administration [117-121]. In addition, lipid nanocarriers have improved the chemical stability of encapsulated drugs as well as reduce the toxicity concerns of administered drugs. This is especially seen in the application of lipid-based nanocarriers e.g. SLN and NLC, in the delivery of drugs with low solubility, bioavailability and very narrow therapeutic index e.g. proteins and peptides, which cannot be administered through the oral route due to enzymatic degradation in the GIT [122]. Research into better and effective means of treatment of parasitic diseases like malaria have employed several lipid-based nanocarriers e.g. liposomes, SLN, NLC, etc for targeted and discriminatory delivery of entrapped antimalarial agent (e.g. chloroquine, primaquine, artemether, etc) to *Plasmodium*-infected Red Blood Cells (RBC) with very low haemolytic potential and increased antimalarial activity [123,124].

Regulatory issues in the use of lipid nanocarriers

Advances made in nanotechnology led to the development and introduction of lipid-based nanodevices and their application for drug delivery for diagnostic, therapeutic, cosmetic, veterinary, and imaging purposes. Consequently, at the dawn of the millennium, formulation scientists have designed, formulated and developed several lipid-based nanocarriers such as SLN, NLC, liposomes, etc for clinical use, and several products made from these devices are already in the market. Since nano-based products are rapidly finding increased applications in almost all aspects of human existence, including medical, military, household, industrial, agricultural, etc, and the increased funding available for research and development of these products, there is a great need for regulation of the development and application of lipid-based nanocarriers so as to guarantee the safety of formulators, consumers as well as the environment. To achieve sensible development and application of these products, many countries have set up and empowered regulatory agencies to oversee the activities of formulators and developers of nano-based products. In the US, the FDA is the agency charged with protecting the public health by assuring the safety, efficacy, and security of human and veterinary drugs, biological products, medical devices, food, cosmetics and radioactive materials. The agency is also responsible for the dissemination of accurate, science-based information about these items and their use for improved health care [125]. Though most of the excipients used in the formulation of lipid nanocarriers are GRAS (generally regarded as safe) materials and are used at will, adequate care should be applied in the use of these ingredients, especially if there is possibility of excipient-drug interaction or when used in high concentrations as they could be toxic. Thus, it is essential to formulate and introduce clear guidelines to be followed before the use of any excipients in lipid-based nanocarriers. Pharmaceutical and allied industries should be encouraged to fund researches in the development of safe excipients from green sources, instead of the current practice of using well-known excipients, as this frustrates and impedes the introduction of new and innovative delivery systems [126]. Formulators in developing countries have formulated lipid nanocarriers using lipids extracted from plant and animal tissues, which have been used in the delivery of several drugs, but there are no set regulations in the extraction and application of these lipid materials for safe drug delivery. It is interesting to note that the FDA maintains an approved inactive Ingredients Guide (IIG) list of excipients used in the production of lipid nanocarriers which specifies the maximum quantity allowed for excipients, which can be used for a specific route of administration. However, the FDA does not have any protocols or procedures through which it evaluates the safety of these excipients separately from products due to the integral roles played by these excipients in formulations [3]. It is therefore important for formulators and other stakeholders actively involved in drug delivery using lipid-based materials to work hard and come up with ideas and science-based information which will be of immense benefit in the drafting of practical guidelines which will be used in all countries of the world for the development and formulation of safe lipid-based nanocarriers.

Future outlook in the use of lipid-based nanocarriers for sustained drug delivery

Lipid-based nanocarriers offer an attractive and highly versatile platform for the delivery of drugs, especially those with solubility and/or bioavailability challenges. Majority of these drugs are classified as class II and IV drugs according to the Biopharmaceutics Classification System (BCS). Till date, the numbers of research groups working with lipid-based nanocarriers as well as the number of publications, patents and products emanating from this specialty have greatly increased. This is an attestation to the wide acceptance by scientists in academia and indus-

tries of the huge potentials of lipid nanosystems as the future of medicine. The documented successes and the accompanying excitement among formulators will not be complete without the pharmaceutical, cosmetic, biomedical and other allied industries making remarkable effort towards the translation of these innovative research findings into tangible products. This is important since most of these companies already have the capacity to produce delivery systems, and so, can easily upgrade their production technology by adapting any of the lipid-based nanocarriers for enhanced production of finished products. Lipid-based nanocarriers have come to stay for a very long time, and it is only right that companies involved in drug delivery accept this latest trend. These nanocarriers offer varying opportunities for sustained drug delivery ranging from oral to parenteral application. Drugs with poor pharmacokinetic and stability profiles have been reformulated and their properties improved for clinical use. Lipid-based carriers can be adapted to address issues relating to oral drug administration such as GIT irritation, instability and unpleasant taste. It is also important to develop technologies using lipid-based nanocarriers to guarantee sustained delivery of proteins, peptides and genetic products via the oral route. Newer and enhanced technologies for the transformation of most of these lipid nanocarriers to tablets, pellets, granules, capsules, etc should be explored, developed and introduced. More research efforts should be committed into the delivery of drugs through the dermal and pulmonary route using lipid-based nanocarriers. There is a lot of work to be done in this regard as the potentials in these areas are yet to be fully tapped. Challenges posed by these delivery routes like crossing the stratus corneum and the brionchioles should be surmounted as this will aid the improved occlusion and permeation of the skin and the delivery of Active Pharmaceutical Ingredients (APIs) into the alveoli for targeted treatment of tissues. Lipid nanocarriers should also be focused on the development of a needs-based strategy for neglected diseases like trypanosomiasis, leishmaniasis, etc which cause tremendous suffering and death, mostly in the poorest countries of the world.

Expert opinion on the use of lipid nanocarriers for sustained drug delivery

Lipid-based nanocarriers can be formulated using feasible and simple technology with overall reduction in production costs. They offer easy and patient-friendly administration. They provide efficient sustained and targeting potentials of encapsulated drugs relative to the conventional, non-lipid dosage forms. Drug delivery using lipid-based nanocarriers modifies the Pharmacokinetic (PK) and Pharmacodynamic (PD) properties of the candidate drug, and opening new vistas for improved efficacy and chemical stability of the nanoparticles. In terms of the technologies for their production, available formulation strategies can be optimized to a large scale for industrial applications. However, challenges encountered in the production and applications of lipid-based nanocarriers such as polymorphic transitions, drug expulsion, gelation, aggregation and co-existing micelles and liposomes, show that a lot of strategies needed to be introduced, reviewed or discarded for the production of quality nanoparticles. In any case, lipid-based nanocarriers appear to be good and interesting devices with a high potential for improved and sustained drug delivery.

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