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SOLID LIPID NANOPARTICLES AS DRUG DELIVERY SYSTEM

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ABSTRACT

Solid lipid nanoparticles (SLNs) are alternative drug delivery system to colloidal drug delivery system such as liposome, lipid emulsions. SLNs are rapidly developing nanotechnology with several applications in drug delivery system, clinical medicine and other science. The ability of SLNs to incorporate drug into nanocarrier that offer new type in drug delivery system. Therefore SLNs is reaching the goal of controlled and site specific drug delivery system. In this article we discussed the preparation method, characterization, route of administration of SLNs, advantages, different preparation method which are suitable for large scale production and application of SLNs. Analytical techniques for characterization of SLNs like photon correlation spectroscopy, scanning electron microscopy, different scanning colorimetry are highlighted.

KEY WORDS

Solid lipid nanoparticles, colloidal drug carriers, nanocarrier, nanotechnology.

INTRODUCTION

Solid lipid nanoparticles (SLNs) are introduced as a carrier system for poorly water soluble drug and cosmetic active drug. Colloidal particles ranging in size between 10 and 1000 nm are known as nanoparticles. They are synthesized from synthetic/natural polymers and suited to optimize drug delivery and reduce toxicity. They have emerged as a variable substitute to liposomes as drug carriers. The successful implementation of nanoparticles for drug delivery depends on their ability to penetrate through several anatomical barriers, sustained release of their contents and their stability in the nanometer size. They have some limitations due to their high cost and scarcity of safe polymers with regulatory approval[1]. To overcome this limitation polymeric nanoparticle lipid is used as an

alternative carrier. These nanoparticles are known as solid lipid nanoparticles (SLNs) [2]. SLNs are developed as an alternative system for polymeric nanoparticles, liposome and emulsion. SLNs have unique property like small size, large surface area, high drug loading and interaction of phase at the interphase[3]. SLNs are attracting major attention in novel colloidal carrier for intravenous application. SLNs are a new generation of submicron-sized lipid emulsions where the liquid lipid (oil) has been substituted by a solid lipid. SLNs are sub-micron colloidal carrier composed of physiological lipid, dispersed in water or in an aqueous surfactant solution[4].

PREPARATION OF SLNs

SLNs are made up of solid lipid, emulsifier and water/solvent. The lipids used may be triglycerides (tri-stearin), partial glycerides (Imwitor), fatty acids (stearic acid, palmitic acid), steroids (cholesterol) and waxes (cetyl palmitate). Various emulsifiers and their combination (Pluronic F 68, F 127) have been used to stabilize the lipid dispersion. The combination of emulsifiers might prevent particle agglomeration more efficiently[3].

PREPARATION METHOD OF SLNs

There are different methods of SLNs preparation like

1. High shear homogenization.
2. Hot homogenization.
3. Cold homogenization.
4. Ultrasonication or high speed homogenization.
5. Micro emulsion based SLN preparations.
6. SLN preparation by using supercritical fluid.
7.SLN prepared by solvent emulsification/ evaporation.
8. Double emulsion method.
9. Spray drying method.

1. High shear homogenization:

High shear homogenization techniques were initially used for the production of solid lipid nanodispersions[5-6]. This method is easy to handle. Dispersion quality is often compromised by the presence of micro particles[7]. Different process parameters, including emulsification time, stirring rate and cooling condition on the particle size and zeta potential are investigated by Olbrich *et al*. Lipids used in this study are tripalmitin, mixture of mono, di, triglycerides(WitepsolW35) with glycerol behenate and poloxamer 188 used as steric stabilizers (0.5% w/w).Witepsol W35 dispersions the best SLN quality was obtained after stirring for 8 min at 20,000 rpm followed by cooling 10 min and stirring at 5000 rpm at a room temp[8]. Higher stirring rates did not significantly change the particle size, but slightly improved the polydispersity index.

2. Hot homogenization:

It is carried out at temperature above the melting point of the lipid and it is similar to homogenization of emulsion. Hot homogenization can be carried out by high pressure homogenizers or high intensity ultrasound. Typically lipid content is between 5-10%. By this method up to 40% success is obtained. A pre-emulsion of the drug loaded lipid melt and the aqueous emulsifier phase (same temperature) is obtained by high-shear mixing device (like silversion-type homogenizer). High pressure homogenization of the pre-emulsion is done above the lipid melting point. The quality of the pre-emulsion affects the quality of the final product to a great extent and it is desirable to obtain droplets in the size range of a few micrometers. Lower particle sizes are obtained at higher processing temperatures due to lower viscosity of the lipid phase[9] and this leads to accelerate drug and carrier degradation. Good product is obtained due to several passes through the high-pressure homogenizer (HPH), typically 3-5 passes. High pressure processing always increases the temperature of the sample[10]. In most cases 3-5 homogenization cycles at 500-1500 bar are sufficient. Increasing the homogenization leads to an increase of the particle size due to particle coalescence which occurs because of the high kinetic energy of the particles.

Problem: High temperature lead to degradation of active compound and metal contamination due to high intensity ultrasound.

3. Cold homogenization:
Cold homogenization has been developed to overcome the following problems of the hot homogenization technique such as: Temperature mediated accelerated degradation of the drug payload, Partitioning and hence loss of drug into the aqueous phase during homogenization[10], Uncertain polymorphic transitions of the lipid due to complexity of the crystallization step of the nanoemulsion lead to several modifications and/or super cooled melts. First step in between cold and hot homogenization is same but they are differing from next steps. The melt containing drug is cooled rapidly using ice or liquid nitrogen for distribution of drug in the lipid matrix. Particle sizes attained are in the range 50-100 microns. Compared to hot homogenization; larger particle sizes and a broader size distribution are typical of cold homogenized samples[11]. Cold homogenization minimizes the thermal exposure of the sample.

4. Ultrasonication or high speed homogenization:
SLNs can also be produce by sonication or high speed stirring[12-13]. It is very simple and it can be advantageous over other method like hot and cold homogenization because the equipment used in this technique is very common in every lab. Disadvantage is like it distributes larger particle size ranging between micrometer range lead to physical instability like particle growth upon storage and also metal contamination due to ultrasonication.

5. Micro emulsion based SLN preparations:
Gasco and coworkers develop a new technique for production of SLNs based on the dilution of micro emulsions[14]. They are made by stirring an optically transparent mixture at 65-70 ^{0} which is typically composed of a low melting fatty acid (stearic acid), an emulsifier (polysorbate 20, polysorbate 60, soy phosphatidylcholine, and sodium taurodeoxycholate), co-emulsifiers (sodium monooctylphosphate) and water. The hot micro emulsion is dispersed in cold water (2-3^0) under stirring. Typical volume ratios of the hot micro emulsion to cold water are in the range of 1:25 to 1:50.The dilution process is critically determined by the composition of the micro emulsion. According to the literature[15-16], the droplet structure is already contained in the micro emulsion and therefore, no energy is required to achieve submicron particle sizes.
According to De Labouret *et al.*[17], the particle size is critically determined by the velocity of the distribution processes. Nanoparticles were produced only with solvents which distribute very rapidly into the aqueous phase (acetone), while larger particle sizes were obtained with more lipophilic solvents. The hydrophilic co-solvents of the micro emulsion might play a similar role in the formation of lipid nanoparticles as the acetone for the formation of polymer nanoparticles[18].

6. SLN preparation by using supercritical fluid:
This is new technique and advantage of solvent-less processing[19-20]. SLN can be prepared by the rapid expansion of supercritical carbon dioxide solutions this method is called as RESS method. Carbon dioxide with 99.99% is good solvent[21].

7.SLN prepared by solvent emulsification/evaporation:
This technique is used for of nanoparticle dispersions by precipitation in o/w emulsions[22]. The lipophilic material is dissolved in water-immiscible organic solvent (cyclohexane) that is emulsified in an aqueous phase. Upon evaporation of the solvent nanoparticle dispersion is formed by precipitation of the lipid in the aqueous medium. The mean diameter of the obtained particles was 25 nm with cholesterol acetate as model drug and lecithin/sodium glycocholate blend as emulsifier. The reproducibility of the result was confirmed by Siekmann and Westesen, who produced the cholesterol acetate nanoparticles of mean size 29 nm[23].

8. Double emulsion method:

Novel method based on solvent emulsification-evaporation has been used for preparation of hydrophilic loaded SLNs[24] .The drug is encapsulated with a stabilizer to prevent drug partitioning to external water phase during solvent evaporation in the external water phase of w/o/w double emulsion.

9. Spray drying method:
It's a cheaper method than lyophilization. This method causes particle aggregation due to high temperature, shear forces and partial melting of the particle. The best result was obtained with SLN concentration of 1% in a solution of trehalose in water or 20% trehalose in ethanol-water mixtures (10/90 v/v) [25].

STERLIZATION OF SLNs

Intravenous and ocular administration SLN must be sterile. The high temperature reach during sterilization by autoclaving causes a hot o/w micro emulsion to form in the autoclave, modifies the size of the hot nanodroplets. On subsequent slow cooling, the SLN reformed, but some nanodroplets may coalesce, producing larger SLN than the initial ones. Amounts of surfactant and co surfactant present in the hot system are smaller. There for the nanoparticles are not stabilized.

ROUTE OF ADMINISTRATION

SLNs are given by following route of administration
1. Oral administration.
2. Parenteral administration.
3. Transdermal application.

1. Oral administration:
Forms of SLNs preparation which are given by oral route are aqueous dispersions. SLNs loaded dosage form such as tablets, pellets and capsule. The microclimate of the stomach favors particle aggregation due to the acidity and high ionic strength. It is to be expected that food will have a large impact on SLN performance[26].

2. Parenteral administration:
SLNs generally administered intravenously to animals. Distribution of SLN were found to have higher drug concentrations in lung, spleen and brain, while the solution led to more distribution into liver and kidneys[27]. SLN showed higher blood levels in comparison to a commercial drug solution after intravenous.

3. Transdermal application:
The smallest particle sizes are observed for SLN dispersions with low lipid content (up to 5%). Disadvantages of dermal administration are low concentration of the dispersed lipid and the low viscosity. The incorporation of the SLN dispersion in an ointment or gel is necessary in order to achieve a formulation which can be administered to the skin[28].

CHARACTERIZATION OF SLNs

Characterization of the SLNs is necessary for its quality control. Characterization of SLN is a serious challenge due to the colloidal size of the particles and the complexity and dynamic nature of the delivery system.
Parameter which are to be evaluated: Particle size, zeta potential, drug release, surface morphology. Polymorphism, degree of crystallinity, time scale of distribution processes.

PARTICLE SIZE AND ZETA POTENTIAL**:**
There are so many techniques for the particle size and zeta potential (size distribution) like photon correlation spectroscopy (PCS), transmission electron microscopy (TEM), scanning electron microscopy (SEM), atomic force microscopy (AFM), scanning tunneling microscopy (STM) or freeze fracture electron microscopy (FFEM).
For the routine measurement of particle size Photon correlation spectroscopy (PCS) and laser diffraction (LD) are important techniques used. Coulter counter are rarely used to measure particle size because of difficulties in the assessment of small nanoparticle. Photon correlation spectroscopy (PCS) is not able to detect larger microparticles. Difficulties may arise both in PCS and LD measurements for samples which contain several populations of different

size[29]. Therefore, additional techniques might be useful like light microscopy it gives fast indication of the presence and character of microparticles. Electron microscopy provides, in contrast to PCS and LD, direct information on the particle shape. However, the investigator should pay special attention to possible artifacts which may be caused by the sample preparation. For example, solvent removal may cause modifications which will influence the particle shape. Zeta potential is an important product characteristic of SLNs since its high value is expected to lead to deaggregation of particles in the absence of other complicating factors such as steric stabilizers or hydrophilic surface appendages. It is usually measured by zetameter.

STATIC LIGHT SCATTERING/FRAUNHOFER DIFFRACTION**:**
The method is fast and rugged, but requires more cleanliness than DLS, and advance knowledge of the particles' optical qualities Static light scattering (SLS) is an ensemble method in which the pattern of light scattered from a solution of particles is collected and fit to fundamental electromagnetic equations in which size is the primary variable.

DYNAMIC LIGHT SCATTERING (DLS):
DLS, also known as PCS or quasi-elastic light scattering (QELS) records the variation in the intensity of scattered light on the microsecond time scale. This variation results from interference of light scattered by individual particles under the influence of *BROWNIAN MOTION* and is quantified by compilation of an autocorrelation function. This function is fit to an exponential, or some combination or modification thereof, with the corresponding decay constant(s) being related to the diffusion coefficient. Using standard assumptions of spherical size, low concentration, and known viscosity of the suspending medium, particle size is calculated from this coefficient.The advantages of the method are the speed of analysis, lack of required calibration, and sensitivity to submicrometer particles[29].

ELECTRON MICROSCOPY:
Scanning electron microscopy (SEM) and transmission electron microscopy (TEM) provide a way to directly observe nanoparticles and physical characterization of nanoparticles. TEM has a smaller size limit of detection, is a good validation for other methods and one must be cognizant of the statistically small sample size and the effect that vacuum can have on the particles[29].
NUCLEAR MAGNETIC RESONANCE (NMR):
NMR is used to determine both size and nature of nanoparticles. The selectivity afforded by chemical shift complements the sensitivity to molecular mobility to provide information on the physicochemical status of components within the nanoparticle.

ATOMIC FORCE MICROSCOPY (AFM):
In this technique, a probe tip with atomic scale sharpness is kept across a sample to produce a topological map based on the forces at play between the tip and the surface. The probe can be dragged across the sample (contact mode) or allowed to hover just above (noncontact mode), with the exact nature of the particular force employed serving to distinguish among the sub techniques[29]. That ultrahigh resolution is obtainable with this approach, which along with the ability to map a sample according to properties in addition to size.

ACOUSTIC METHODS:
Acoustic spectroscopy measures the attenuation of sound waves as a means of determining size through the fitting of physically relevant equations. In addition, the oscillating electric field generated by the movement of charged particles under the influence of acoustic energy can be detected to provide information on surface charge.

X-RAY DIFFRACTION AND DIFFERENTIAL SCANNING CALORIMETRY (DSC):
The geometric scattering of radiation from crystal planes within a solid allow the presence or absence of the former to be determined thus permitting the degree of crystallinity to be

assessed. DSC can be used to determine the nature and speciation of crystallinity within nanoparticles through the measurement of glass and melting point temperatures and their associated enthalpies[30].

ADVANTAGE:

1. SLNs have better stability and ease of upgradability to production scale as compared to liposome.
2. In SLNs the lipid matrix is made from physiological lipid which decreases the danger of acute and chronic toxicity[3].
3. Very high long-term stability.
4. It is easy to manufacture than bipolymeric nanoparticles.
5. Better control over release kinetics of encapsulated compound.
6. SLNs can be enhancing the bioavailability of entrapped bioactive.
7. Chemical protection of labile incorporated compound.
8. Raw material which are to be required are same as that of emulsion.
9. Large scale production is possible.
10. High concentration of functional compound can be achieved.
11. Lyophilization possible.

DISADVANTAGE:

1. Poor drug loading capacity[31].
2. Drug expulsion after polymeric transition during storage[31].
3. Relatively high water content of the dispersions (70-99.9%) [32].
4. The low capacity to load hydrophilic drugs due to partitioning effects during the production process[33].

APPLICATION

1. Oral SLNs in ant tubercular chemotherapy:
Antitubercular drugs such as rifampicin, isonizide, pyrazinamide-loaded SLN systems, were able to decrease the dosing frequency and improve patient compliance[34]. By using the emulsion solvent diffusion technique this antitubercular drug loaded solid lipid nanoparticles are prepared.

2. SLNs for topical use:
SLNs used for topical application for various drug such as anticancers[35], vitamin-A[36], isotretinoin, flurbiprofen[37].Using glyceryl behenate, vitamine A-loaded nanoparticles can be prepared. This method is useful for the improvement of penetration with sustained release. The isotretinoin-loaded lipid nanoparticles was formulated for topical delivery of drug.Production of the flurbiprofen-loaded SLN gel for topical application offer a potential advantage of delivering the drug directly to the site of action, which will produce higher tissue concentrations.

3. SLNs as cosmeceuticals:
The SLNs have been applied in the preparation of sunscreens and as an active carrier agent for molecular sunscreens and UV blockers[38]. SLN and NLCs have proved to be controlled release innovative occlusive topicals. Better localization has been achieved for vitamin A in upper layers of skin with glyceryl behenate SLNs compared to conventional formulations.

4. SLNs as gene vector carrier:
SLN can be used in the gene vector formulation[39]. There are several recent reports of SLN carrying genetic/peptide materials such as DNA, plasmid DNA and other nucleic acids[40]. The gene transfer was optimized by incorporation of a diametric HIV-1 HAT peptide (TAT 2) into SLN gene vector. The lipid nuclic acid nanoparticles were prepared from a liquid nanophase containing water and a water miscible organic solvent where both lipid and DNA are separately dissolved by removing the organic solvent, stable and homogeneously sized lipid-nuclic acid nanoparticle (70-100 nm) were formed. It's called genospheres. It is targeted specific by insertion of an antibody-lipo polymer conjugated in the particle.

5. SLNs in breast cancer and lymph node metastases:

Mitoxantrone-loaded SLN local injections were formulated to reduce the toxicity and improve the safety and bioavailability of drug[41]. Efficacy of doxorubicin (Dox) has been reported to be enhanced by incorporation in SLNs.In the methodology the Dox was complexed with soybean-oil-based anionic polymer and dispersed together with a lipid in water to form Dox-loaded solid lipid nanoparticles The system has enhanced its efficacy and reduced breast cancer cells.

6. SLNs as a targeted carrier for anticancer drug to solid tumors:

SLNs have been reported to be useful as drug carriers to treat neoplasms[42]. Tumour targeting has been achieved with SLNs loaded with drugs like methotrexate[43] and camptothecin[44]. Tamoxifen a anticancer drug is incorporated in SLN to prolong release of drug after i.v.

7. Stealth nanoparticles:

These provide a novel and unique drug-delivery system they evade quick clearance by the immune system. Such nanoparticles can target specific cells. Stealth SLNs have been successfully tested in animal models with marker molecules and drugs. Antibody labelled stealth lipobodies have shown increased delivery to the target tissue in accessible sites[45].

8. SLNs for potential agriculture application:

Essential oil extracted from *Artemisia arboreseens* L when incorporated in SLN, were able to reduce the rapid evaporation compared with emulsions and the systems have been used in agriculture as a suitable carrier of ecologically safe pesticides[46].

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