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Formulation of solid lipid nanoparticles and their applications.

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Abstract
Solid lipid nanoparticles (SLN) introduced in 1991 represent an alternative carrier system to traditional colloidal carriers, such as emulsions, liposomes and polymeric micro- and nanoparticles. SLN are aqueous colloidal dispersions, the matrix of which comprises of solid biodegradable lipids. SLN are manufactured by techniques like high pressure homogenization, solvent diffusion method etc. SLN combine advantages of the traditional systems but avoid some of their major disadvantages. They exhibit major advantages such as modulated release, improved bioavailability, protection of chemically labile molecules like retinol, peptides from degradation, cost effective excipients, improved drug incorporation and wide application spectrum. However there are certain limitations associated with SLN, like limited drug loading capacity and drug expulsion during storage, which can be minimized by the next generation of solid lipids, Nanostructured lipid carriers (NLC). NLC are lipid particles with a controlled nanostructure that improves drug loading and firmly incorporates the drug during storage. Due to their unique size-dependent properties, lipid nanoparticles offer the possibility to develop new therapeutics. Owing to their properties and advantages, SLN and NLC may find extensive application in topical drug delivery, oral and parenteral administration of cosmetic and pharmaceutical actives. Cosmeceuticals is emerging as the biggest application target of these carriers. Carrier systems like SLN and NLC were developed with a perspective to meet industrial needs like scale up, qualification and validation, simple technology, low cost etc. The ability to incorporate drugs into nanocarriers offers a new prototype in drug delivery that could be used for secondary and tertiary levels of drug targeting. This review mainly focuses on the advantages and limitations of the solid lipid nanoparticles over other colloidal carriers and different techniques available for the formulation of SLNs and their applications in therapeutics.

Key Words
Solid lipid nanoparticles (SLN), colloidal drug carriers, homogenization.

Introduction
Nanoparticles are subnanosized colloidal structures composed of synthetic or semi-synthetic polymers ranging in size between 10 and 1000 nm. The solid lipid nanoparticles are sub micron colloidal carriers (50-100 nm) which are composed of physiological lipid, dispersed in water or in aqueous surfactant solution. SLNs as colloidal drug carrier combines the advantage of polymeric nanoparticles fat emulsions and liposomes. In order to overcome the disadvantages associated with the liquid state of the oil droplets, the liquid lipid was replaced by solid lipid which eventually transformed into solid lipid nanoparticles.[1,2]

Advantages of SLN [2,3]

- Small size and relatively narrow size distribution which provide biological opportunities for site-specific drug delivery by SLNs.
- Controlled release of active drug over a long period can be achieved.
- Protection of incorporated drug against chemical degradation.
- Possible sterilization by autoclaving or gamma irradiation.
- SLNs can be lyophilized as well as spray dried.
- No toxic metabolites are produced.
- Avoidance of organic solvents.
- Relatively cheaper and stable.
- Ease of industrial scale production by hot dispersion technique.
- Incorporation of drug can reduce distinct side effects of drug, e.g. Thrombophlebitis that is associated with i.v. injection of diazepam.

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- Surface modification can easily be accomplished and hence can be used for site-specific drug delivery system.

SLN versus Other colloidal carriers[4,5]

SLN have been proven to be a better alternative carrier system than conventional 0/w emulsion in the following aspects.

- If protection of drug against chemical degradation is required. Incorporation of drug in the solid lipid matrix surely offer a better protection than can be achieved in the oily internal phase of emulsion and liposomes
- Prolonged release of drug from emulsion is not feasible which can be achieved to a certain extent from SLN

SLNs is found to be a better carrier than polymeric nanoparticles in the following aspects

- Lower cytotoxicity due to the absence of solvents
- Low cost of excipients
- Large scale production is possible by the simple process of high-pressure homogenization

SLNs versus liposomes

- In comparison with liposomes SLNs offer better protection to drug against chemical degradation there is no or little access of water to the inner core of lipid particles
- Depending upon the nature of the drug higher payload might be achieved.

Nanostructured lipid carriers (NLC):[6,7,8]

NLC were introduced to overcome the potential difficulties with SLNs. The goal was to increase the drug loading and prevent drug expulsion. This could be visualized in three ways. In the first model, spatially different lipids (like glycerides) composed of different fatty acids are mixed. The use of spatially different lipids leads to larger distances between the fatty acid chains of the glycerides and general imperfections in the crystal and thus provides more room for accommodation of guest molecules. The highest drug load could be achieved by mixing solid lipids with small amounts of liquid lipids (oils). This model is called imperfect type NLC. Drugs showing higher solubility in oils than in solid lipids can be dissolved in the oil and yet be protected from degradation by the surrounding solid lipids. These types of NLC are called multiple types NLC, and are analogous to w/o/w emulsions since it is an oil-in-solid lipid-in-water dispersion.[9,10]

Lipid drug conjugates (LDC):[11,12]

A major problem of SLNs is the low capacity to load hydrophilic drugs due to partitioning effects during the production process. Only highly potent low dose hydrophilic drugs may be suitably incorporated in the solid lipid matrix. In order to overcome this limitation, the so called LDC nanoparticles with drug loading capacities of up to 33% have been developed. An insoluble drug- lipid conjugate bulk is first prepared either by salt formation (e.g. with a fatty acid) or by covalent linking (e.g. to ester or ethers). The obtained LDC is then processed with an aqueous surfactant solution (such as Tweens) to a nanoparticle formulation using high pressure homogenization (HPH). Such matrices may have potential application in brain targeting of hydrophilic drugs in serious protozoal infections.

Preparation methods of SLNs

Hot homogenization technique

The hot homogenization technique can be applied to lipophilic and insoluble drugs. Many heat sensitive drugs can be safely processed because the exposure time to high temperature is relatively short. The technique does not suit for incorporation of hydrophilic drugs into SLN because of higher partition of drug in water during homogenization those results in low entrapment efficiency.[13,14]

Cold Homogenization technique

For hydrophilic drugs, the cold homogenization technique is the method of first choice. In case of poor solubility of the hydrophilic drug in the melted lipid, surfactants can be used for solubilization of the drug. This homogenization technique avoids and minimizes melting process of lipid and hence it is suitable for thermo sensitive and thermolabile drugs.

Ultrasonication or high speed homogenization[15,16]

SLN were also developed by high speed stirring or sonication. A most advantages is that, equipment whatever use here are very common in every lab. The problem of this method is broader particle size distribution ranging into micrometer range. This lead physical instabilities likes particle growth upon storage. Potential metal contamination due to ultrasonication is also a big problem in this method. So for making a stable formulation, studies have been performed by various research groups that high speed stirring and ultrasonication are used combined and performed at high temperature.

SLN prepared by solvent emulsification/evaporation

For the production of nanoparticle dispersions by precipitation in o/w emulsions 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.

Micro emulsion based SLN preparations

Gasco and co-workers developed SLN preparation techniques which are based on the dilution of microemulsions. They are made by stirring an optically transparent mixture at 65-700 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 microemulsion is dispersed in cold water (2-30) under stirring. Typical volume ratios of the hot microemulsion to cold water are in the range of 1:25 to 1:50. The dilution process is critically determined by the composition of the microemulsion. According to the literature, the droplet structure is already contained in the microemulsion and therefore, no energy is required to achieve submicron particle sizes. With respect to the similarities of the production procedure of polymer nanoparticles described by French scientists, different mechanisms might be considered. Fessi produced polymer particles by dilution of polymer solutions in water. According to De Labouret *et al* 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 microemulsion might play a similar role in the formation of lipid nanoparticles as the acetone for the formation of polymer nanoparticles.

SLN preparation by using supercritical fluid[17]

This is a relatively new technique for SLN production and has the advantage of solvent-less processing. There are several variations in this platform technology for powder and nanoparticle preparation. SLN can be prepared by the rapid expansion of supercritical carbon dioxide solutions (RESS) method. Carbon dioxide (99.99%) was the good choice as a solvent for this method.[18]

Spray drying method[19]

It's an alternative procedure to lyophilization in order to transform an aqueous SLN dispersion into a drug product. It's a cheaper method than lyophilization. This method cause particle aggregation due to high temperature, shear forces and partial melting of the particle. Freitas and Mullera recommends the use of lipid with melting point >70 0 for spray drying. 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).

Double emulsion method

For the preparation of hydrophilic loaded SLN, a novel method based on solvent emulsification-evaporation has been used. Here 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.

Characterization of solid lipid nanoparticles[20]

Adequate and proper characterization of the SLNs is necessary for its quality control. However, 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. The important parameters which need to be evaluated for the SLNs are, particle size, size distribution kinetics (zeta potential), degree of crystallinity and lipid modification (polymorphism), coexistence of additional colloidal structures (micelles, liposome, super cooled, melts, drug nanoparticles), time scale of distribution processes, drug content, *in vitro* drug release and surface morphology.

Measurement of particle size and zeta potential[37]

Photon correlation spectroscopy (PCS) and laser diffraction (LD) are the most powerful techniques for routine measurements of particle size. The Coulter method is rarely used to measure SLN particle size because of difficulties in the assessment

of small nanoparticle and the need of electrolytes which may destabilize colloidal dispersions. PCS (also known dynamic light scattering) measures the fluctuation of the intensity of the scattered light which is caused by the particle movement. This method covers a size range from a few nanometers to about 3 microns. This means that PCS is a good tool to characterize nanoparticles, but it is not able to detect larger microparticles. They can be visualized by means of LD measurements. This method is based on the dependence of the diffraction angle on the particle radius (Fraunhofer spectra). Smaller particles cause more intense scattering at high angles compared to the larger ones. A clear advantage of LD is the coverage of a broad size range from the nanometer to the lower millimeter range.

Degree of crystallinity

It can be measured by X-ray diffraction (powder X-ray diffraction) .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. Another method that is a little different from its implementation with bulk materials, 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.

Applications[21,22]

Solid lipid Nanoparticles possesses a better stability and ease of upgradability to production scale as compared to liposomes. This property may be very important for many modes of targeting. SLNs form the basis of colloidal drug delivery systems, which are biodegradable and capable of being stored for at least one year. They can deliver drugs to the liver *in vivo* and *in vitro* to cells which are actively phagocytic. There are several potential applications of SLNs some of which are given below.

SLNs as gene vector carrier:[23,24]

SLN can be used in the gene vector formulation. In one work, the gene transfer was optimized by incorporation of a diametric HIV-1 HAT peptide (TAT 2) into SLN gene vector. There are several recent reports of SLN carrying genetic/peptide materials such as DNA, plasmid DNA and other nucleic acid. 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 17 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.

SLNs for topical use[25,26,27,28]

SLNs and NLCs have been used for topical application for various drugs such as tropolide, imidazole, antifungals, anticancers, vitamin A,isotretinoin, ketoconazole, DNA, flurbiprofen and glucocorticoids. The penetration of podophyllotoxin-SLN into stratum corneum along with skin surface lead to the epidermal targeting. By using glyceryl behenate, vitamine A-loaded nanoparticles can be prepared. The methods are useful for the improvement of penetration with sustained release. The isotretinoin-loaded lipid nanoparticles was formulated for topical delivery of drug. The soyabean lecithin and Tween 80 are used for the hot homogenization method for this. The methodology is useful because of the increase of accumulative uptake of isotretinoin in skin. Production of the flurbiprofen-loaded SLN gel for topical application after a potential advantage of delivering the drug directly to the site of action, which will produce higher tissue concentrations. Polyacrylamide, glycerol and water were used for the preparation of this type of SLN gel.[30,31]

SLNs as cosmeceuticals[32]

The SLNs have been applied in the preparation of sunscreens and as an active carrier agent for molecular sunscreens and UV blockers [54]. The *in vivo* study showed that skin hydration will be increased by 31% after 4 weeks by addition of 4% SLN to a conventional cream.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.

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 pesticide. The SLN were prepared here by using compritol 888 ATO as lipid and poloxamer 188 or Miranol Ultra C32 as surfactant.

SLNs as a targeted carrier for anticancer drug to solid tumors[33,34]

SLNs have been reported to be useful as drug carriers to treat neoplasms. Tamoxifen, an anticancer drug incorporated in SLN to prolong release of drug after i.v. administration in breast cancer and to enhance the permeability and retention effect. Tumour targeting has been achieved with SLNs loaded with drugs like methotrexate and camptothecin.

SLNs in breast cancer and lymph node metastases[35]

Mitoxantrone-loaded SLN local injections were formulated to reduce the toxicity and improve the safety and bioavailability of drug. 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 is enhanced its efficacy and reduced breast cancer cells.[36]

Oral SLNs in antitubercular chemotherapy[37]

Antitubercular drugs such as rifampicin, isonizide, pyrazinamide-loaded SLN systems, were able to decrease the dosing frequency and improve patient compliance. By using the emulsion solvent diffusion technique this anti tubercular drug loaded solid lipid nanoparticles are prepared. The nebulization in animal by incorporating the above drug in SLN also reported for improving the bioavailability of the drug.

Stealth nanoparticles[38,39]

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

Conclusions

SLN as colloidal drug carrier combines the advantage of polymeric nanoparticles, fat emulsions and liposomes. SLNs are prepared by hot and cold homogenization technique and by various advanced techniques. The site specific and sustained release effect of drug can better achieved by using SLNs. A major problem of SLNs is the low capacity to load hydrophilic drugs due to partitioning effects during the production process. Only highly potent low dose hydrophilic drugs may be suitably incorporated in the solid lipid matrix by using NLC drug loading can be improved and drug expulsion can be prevented. By using LDC nanoparticles with drug loading capacities of up to 33% can be achieved. They are having wide applications in therapeutics. So in future research can be carried out on formulation of NLC and LDC to achieve better drug loading, site specificity and low toxic effects.

References

1. Muller RH, Runge SA. Solid lipid nanoparticles (SLN) for controlled drug delivery, Benita (Ed.), Submicron emulsions in drug targeting and delivery, Amsterdam: Harwood Academic Publishers; 1998, p. 219-34.
2. Jenning V, Gysler A, Schafer-Korting M, Gohla S. Vitamin A loaded solid lipid nanoparticles for topical use: occlusive properties and drug targeting to the upper skin. Eur J Pharm Biopharm 2000, 49,211-8.
3. Muller RH, Radtke M, Wissing SA. Nanostructured lipid matrices for improved microencapsulation of drug. Int J Pharm 2002, 242:121-8.
4. Radtke M, Muller RH. Comparison of structural properties of solid lipid nanoparticles (SLN) versus other lipid particles. Proc Int Symp Control Rel Bioact Mater 2000; 27:309-10.
5. Uner M. Preparation, characterization and physico-chemical properties of solid lipid nanoparticles (SLN) and nanostructured lipid carriers (NLC): their benefits as colloidal drug carrier systems. Pharmazie 2006, 61,375-86.
6. Muller RH, Radtke M, Wissing SA. Solid lipid nanoparticles (SLN) and nanostructured lipid carriers (NLC) in cosmetic and dermatological preparations. Adv Drug Deliv Rev, 2002; 54:S131-55.
7. Souto EB, Wissing SA, Barbosa CM, Muller RH. Development of a controlled release formulation based on SLN and NLC for topical clotrimazole delivery. Int J Pharm, 2004; 278:71-7.
8. Souto EB, Muller RH. Investigation of the factors influencing the incorporation of clotrimazole in SLN and NLC prepared by hot high-pressure homogenization. J Microencapsul, 2006; 23:377-88.

9. Souto EB, Muller RH. SLN and NLC for topical delivery of ketoconazole. J Microencapsul 2005; 22:501-10.
10. Souto EB, Muller RH. The use of SLN and NLC as topical particulate carriers for imidazole antifungal agents. Pharmazie 2006; 61:431-37.
11. Morel S, Terreno E, Ugazio E, Aime S, Gasco MR. NMR relaxometric investigations of lipid nanoparticles (SLN) containing gadolinium (III) complexes. Eur J Pharm Biopharm, 1998; 45:157-63.
12. Olbrich C, Gebner A, Kayser O, Muller RH. Lipid–drug conjugate (LDC) nanoparticles as novel carrier system for the hydrophilic antitrypanosomal drug diminazenediaceturate. J Drug target 2002, 10,387-96.
13. Ahlin P, Kristl J, Kobar S. Optimization of procedure parameters and physical stability of solid lipid nanoparticles in dispersion. Acta Pharm 1998; 48:257-67.
14. Jahnke S. The theory of high pressure homogenization, in: Muller RH, Benita S, Bohm B. editors. Emulsions and nanosuspensions for the formulation of poorly soluble drugs, Stuttgart Medpharm Scientific Publishers; 1998; p. 177-2005.
15. Siekmann B, Westesen K. Investigations on solid lipid nanoparticles prepared by precipitation in o/w emulsions. Eur J Pharm Biopharm 1996; 43:104-9.
16. Gasco MR. Method for producing solid lipid microspheres having narrow size distribution. United State Patent. USS 188837; 1993.
17. Cavalli R, Marengo E, Rodriguez L, Gasco MR. Effects of some experimental factors on the production process of solid lipid nanoparticles. Eur J Pharm Biopharm 1996; 43:110-5.
18. Chen YJ, Jin RX, Zhou YQ, Zeng J, Zhang H, Feng QR. Preparation of solid lipid nanoparticles loaded with Xionggui powder-supercritical carbon dioxide fluid extraction and their evaluation in vitro release. Zhongguo Zhong Yao Za Zhi 2006; 31:376-9.
19. Freitas C, Mullera RH. Spray-drying of Solid lipid nanoparticles (SLN TM). Eur J Pharm Biopharm 1998; 46:145-51.
20. Yang S, Zhu J, Lu Y, Ling B, Yang C. Body distribution of camptothecin solid lipid nanoparticles after oral administration. Pharm Res 1999; 16:751-7.
21. Yang SC, Lu LF, Cai Y, Zhu JB, Liang BW, Yang CZ. Body distribution in mice of intravenously injected camptothec in solid lipid nanoparticles and targeting effect on brain. J Control Release 1999; 59:299-307.
22. Zur Muhlen A, Mehnert W. Drug release and release mechanism of prednisolone loaded Solid lipid nanoparticles. Pharmazie 1998; 53:552-5.
23. Rudolph C, Schillinger U, Ortiz A, Tabatt K, Plank C, Muller RH, *et al*. Application of novel Solid lipid nanoparticles (SLN)- gene vector formulations based on a diametric HIV-1 VAT – peptide in vitro and in vivo. Pharmaceutic Res 2004; 21(9):1662-9.
24. Pedersen N, Hansen S, Heydenreich AV, Kristensen HG, Poulsen HS. Solid lipid nanoparticles can effectively bind DNA, streptavidin and biotinylated ligands. Eur J Pharm Biopharm, 2006; 62:155-62.
25. Mei Z, Wu Q. Triptolide loaded solid lipid nanoparticle hydrogel for topical application Drug Develop Ind Pharm 2005; 31:161-8.
26. Chen H, Chang X, Du D, Liu W, Liu J. Weng T, *et al*. Podophyllotoxin-loaded solid lipid nanoparticles for epidermal targeting. J Control Release 2006; 110:296-306.
27. Jenning V, Schafer-Korting M, Gohla S. Vitamin A-loaded solid lipid nanoparticles for topicaluse: drug release properties. J Control Release 2000; 66:115-26.
28. Liu J, Hu W, Chen H, Ni Q, Xu H, Yang X. Isotretinoin-loaded solid lipid nanoparticles with skin targeting for topical delivery. Int J Pharm 2007; 328:191-5.
29. Jain SK, Chourasia MK, Masuriha R. Solid lipid nanoparticles bearing flurbiprofen for transdermal delivery. Drug Deliv., 2005; 12:207-15.
30. Santos MC, Mehnert W, Schaller M. Drug targeting by solid lipid nanoparticles for dermal use.J Drug Target 2002;10:489-95.
31. Mei Z, Wu Q. Triptolide loaded solid lipid nanoparticle hydrogel for topical application. Drug Develop Ind Pharm, 2005; 31:161-8.
32. Wissing S, Lippacher A, Muller R. Investigations on the occlusive properties of solid lipid nanoparticles (SLN). J Cosmet Sci 2001; 52:313-24.
33. Murthy RSR. Solid lipid nanoparticles as carriers for anti-cancer drugs to solid tumours. Drug Deliv 2005; 12:385-92.
34. Ruckmani K, Sivakumar M, Ganeshkumar PA. Methotrexate loaded solid lipid nanoparticles (SLN) for effective treatment of carcinoma. J Nanosci Nanotechnol 2006; 6:2991-5.
35. Lu B, Xiong SB, Yang H., Yin XD, Chao RB. Solid lipid nanoparticles of mitoxantrone for local injection against breast cancer and its lymphnode metastases. Eur J Pharm Sci, 2006; 28:86-95.

36. Wong HL, Rauth AM, Bendayan R. A new polymer-lipid hybrid nanoparticle system increases cytotoxicity of doxorubicin against multidrug-resistant human breast cancer cells. Pharm Res, 2006; 23:1574-85.
37. Pandey R, Sharma S, Khuller GK. Oral SLN Based antitubercular chemotherapy. Tuberculosis (Edinb). 2005; 85:415-20.
38. Dvorak HF, Wagy AJ, Dvorak JT, Dvorak AM. Stealth liposomes. J Pathol 1998; 133:95-108.
39. Wang Y, Wu W. In situ evading of phagocytic uptake of stealth solid lipid nanoparticles by mouse peritoneal macrophages. Drug Deliv 2006; 13:189-92.

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