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Nanostructured lipid matrices for improved microencapsulation of drugs

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Abstract

At the beginning of the nineties solid lipid nanoparticles (SLN) have been introduced as a novel nanoparticulate delivery system produced from solid lipids. Potential problems associated with SLN such as limited drug loading capacity, adjustment of drug release profile and potential drug expulsion during storage are avoided or minimised by the new generation, the nanostructured lipid carriers (NLC). NLC are produced by mixing solid lipids with spatially incompatible lipids leading to special structures of the lipid matrix, i.e. three types of NLC: (I) the imperfect structured type, (II) the structureless type and (III) the multiple type. A special preparation process–applicable to NLC but also SLN–allows the production of highly concentrated particle dispersions (>30–95%). Potential applications as drug delivery system are described.

Keywords: Solid lipid nanoparticles; Controlled release; Nanostructure; Stability

1. Introduction

The use of solid lipid matrices for the prolonged release of drugs is known in pharmacy for many years, i.e. drug release from lipid pellets. Based on this, consequently it was only a matter of time until the production of lipid microparticles, e.g. by spray-congealing by Speiser et al. (Eldem et al., 1991). In the next step the first generation of lipid nanoparticles was produced, the so-called lipid nanopellets for oral administration (Speiser, 1990). For certain reasons this development was not continued. At the beginning of the nineties the second generation was developed, the solid lipid nanoparticles (SLN) either produced by high pressure homogenisation (Müller and Lucks, 1996) or alternatively using the microemulsion precipitation technique (Gasco, 1993). A detailed review of the state of the art of the SLN is given in recently published reviews (Müller et al., 2000; Mehnert and Mäder, 2001).

The development of SLN overcame many problems related to 'traditional' nanoparticulate car-

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rier technologies which limited the use (e.g. liposomes, i.v. emulsions) or even prevented the introduction to the market (e.g. polymeric nanoparticles). SLN can be prepared from regulatorily accepted excipients, these excipients are well tolerated, large scale production by high pressure homogenisation is possible – even using existing production lines for i.v. emulsions, and the technology and the product itself are low cost. However, there are also some potential limitations. Especially the review by Mäder and Mehnert critically highlights potential limitations which might occur:

1. limitation in drug loading capacity;
2. drug expulsion during storage;
3. high water content of aqueous SLN dispersions (70–95%).

This consequently led to a new, improved generation of lipid nanoparticles, the NLC. In contrast to SLN being produced from solid lipids, the NLC are produced by controlled mixing of solid lipids with spatially incompatible liquid lipids leading to special nanostructures with improved drug incorporation and release properties. The paper describes the different types of NLC including production of highly concentrated particle dispersions and their application as drug delivery system.

2. Materials and methods

The lipids (e.g. Imwitor 900) were obtained from Condea (Witten, Germany) or alternatively (e.g. Compritol 888ATO, Precifac ATO) from Gattefossé (Weil am Rhein, Germany). The surfactants Miranol etc. were obtained from Rhodia (Frankfurt, Germany). All other materials were purchased from Sigma Chemicals (Deisenhofen, Germany).

Particle production was performed using a Micron LAB 40 (APV Systems GmbH, Unna, Germany). Typically particle production was performed at 500 bar applying different numbers of production cycles. The stirring process was performed using an Ultra-Turrax T25 (Janke & Kunkel, Staufen, Germany).

3. Results and discussion

3.1. Types of NLC

Since many years it is well known from suppositories that drug expulsion can occur during storage. The lipid transforms to the more perfect β-modification, the increase in perfection of the crystal leaves less space to accommodate drug molecules thus leading to drug expulsion. The same phenomenon can potentially occur when producing SLN.

SLN are produced by dissolving or dispersing the drug in the lipid melt, the lipid melt is dispersed in a hot aqueous surfactant mixture by high speed stirring and the obtained pre-emulsion homogenised at temperatures above the melting point of the lipid (hot homogenisation method (Müller and Lucks, 1996). Alternatively a lipid microemulsion containing the drug is prepared, subsequently the microemulsion poured into cold water to precipitate the lipid nanoparticles (Gasco, 1993). Crystallisation behaviour is different on nanoscale, the solidification temperature is decreased compared with bulk lipid (Bunjes, 1998), in addition the formation of less ordered lipid modification is more pronounced compared with bulk ware (Freitas and Müller, 1999). However, during storage (sometimes within days or hours) more perfect crystalline β-modifications can be formed leading to drug expulsion and drug crystals in the aqueous SLN dispersion.

In addition, formation of more or highly ordered lipid crystals limits of course the drug loading capacity. Perfect lipid crystals can be formed when the lipid molecules are chemically identical, e.g. using highly purified tristearine for lipid particle production (Bunjes et al., 1996).

Therefore lipid particles were developed providing a special nanostructure with better drug accommodation facility, the so-called NLC. A prerequisite for good drug accommodation are larger distances between fatty acid chains of the glycerides and general imperfections in the crystal (e.g. to accommodate amorphous drug clusters). Distances between fatty acid chains can be increased by using glycerides being composed of very different fatty acids (e.g. in length of

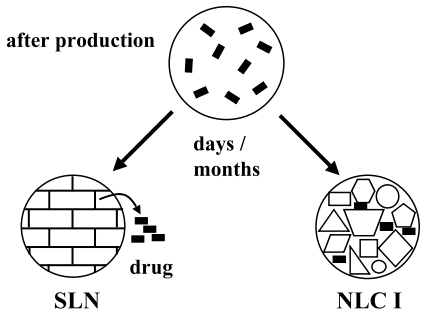

Fig. 1. Crystallisation process during storage to perfect crystal in SLN (left) and unchanged remaining NLC I structure with imperfections (right).

C chain, mixture of saturated and unsaturated acids). To achieve 'highest incompatibility', instead of taking a solid lipid as for SLN, the novel lipid particle was produced by mixing solid lipids with chemically very different liquid lipids (oils). This leads to more imperfections in the crystal and higher drug load (Fig. 1, right).

The fact that the crystallisation process itself causes expulsion of the drug led to the concept of producing NLC which are solid but not crystalline. By using special mixtures of solid lipids and liquid lipids (e.g. hydroxyoctacosanylhydroxystearate, isopropylmyristate) the particles become solid after cooling but do not crystallise (Fig. 2). The solid state (reduction in mobility of lipids) could be verified by NMR measurements, the absence of a melting process by DSC measurements (Müller and Jenning, 1999; Jenning, 1999).

In general the solubility of drugs is higher in liquid lipids (oils) compared with solid lipids. This has to be considered when producing SLN, the

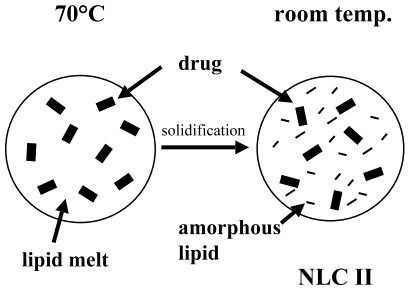

Fig. 2. Structureless type II of NLC—the lipid solidifies in the solid but amorphous state.

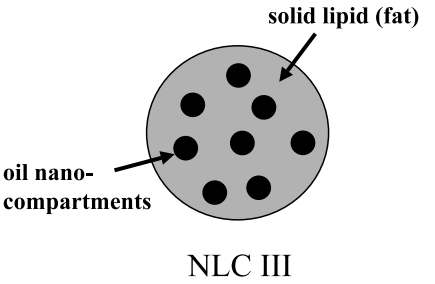

Fig. 3. Theoretical proposed structure of multiple type NLC (oil-in-solid fat-in-water, O/F/W).

drug amount soluble in the lipid melt before particle production is higher than in the final solid lipid nanoparticle. Too high drug concentration in the melt might lead to immediate drug expulsion during the cooling process or dilution in the cold water. Based on this, the multiple type III of NLC was developed. The solid matrix of the lipid nanoparticle contains tiny liquid nanocompartments of oil. In these oil compartments the drug solubility is higher, thus increasing the total drug loading capacity (Fig. 3). The nanocompartments are surrounded by solid lipid matrix, thus still allowing prolonged drug release.

Such small nanocompartments within a nanoparticle (e.g. 200 nm) cannot be created by mechanical means, they are generated by a phase separation process during particle production utilising a miscibility gap. For example, Compritol is mixed with a higher content of Miglyol 812 (e.g. $> 30\%$) and this mixture used for particle production by the hot homogenisation method. In the hot state the two lipids form one phase. During the cooling process phase separation occurs leading to the precipitation of small Miglyol droplets. The presence of free Miglyol could be shown by the crystallisation peaks in DSC (Jenning, 1999). In addition, NMR and DSC measurements confirmed that there was no presence of separate solid lipid particles and oil droplets, the oil compartments are associated with the solid lipid matrix (Zimmermann et al., 2002).

To summarise, there are three different types of NLC:

1. the imperfect type;
2. the structureless type;
3. the multiple O/F/W type.

3.2. Drug incorporation

The first compound formulated in NLC was Retinol. The loading capacity of Retinol in SLN produced from Compritol was only approximately 1%. Producing NLC from a mixture of Compritol and Miglyol 812 increased the loading capacity to 5% (Jenning, 1999). Cyclosporine is also a very attractive molecule for incorporation into SLN. In a previous development, 20% (calculated on the lipid) of cyclosporine could be incorporated into Imwitor 900 SLN (Müller et al., 1998; Penkler et al., 1999) reaching a total cyclosporine concentration of 2%. In the aqueous SLN dispersion (10% lipid) higher drug incorporation led to drug expulsion, formation of cyclosporine crystals in the aqueous suspension. In addition, the 20% cyclosporine SLN had to be lyophilised to avoid drug expulsion during storage.

Cyclosporine-loaded NLC were produced by admixing different oils to solid lipids. It was also possible to create semisolid NLC formulations containing a total of 4% cyclosporine. The corresponding SLN-formulation (only Compritol 888 ATO as lipid) showed the formation of cyclosporine crystals after 8 months of storage at 4 °C. The NLC-batches which additionally contained 12.5 or 25% 2-octyl dodecanol and the same amount of cyclosporine showed less or no crystal formation, respectively, proving a higher loading capacity with increasing amount of liquid lipid (Radtke and Müller 2001; Radtke, 2002).

The phenomenon of burst release of the drug is described for a number of SLN formulations. The burst release is attributed to a phase separation effect. During the cooling process the lipid crystallises first as a drug-free core leading to a drug-enriched outer shell (zur Mühlen et al., 1998; Fig. 4). However, prolonged release is also possible as described by many examples in the literature (Yang et al., 1999; Zara et al., 1999; Jenning et al., 2000). To achieve prolonged release, special production conditions such as low surfactant content and low temperature (e.g. cold homogenisation) are recommended (zur Mühlen and Mehnert, 1998). However, despite applying such production conditions for some drug and lipid mixtures (e.g. etomidate and lipid) the burst release could not be avoided (Radtke, 2002). Applying the NLC technology—especially the imperfect type and amorphous type—provides much more flexibility to achieve the desired prolonged release profile.

3.3. Production technology

The SLN patent (Müller and Lucks, 1996) describes the production of SLN homogenising lipid concentrations between 0.5% up to a maximum of 30%. Homogenisation of higher concentrations led to ointments which were believed to be of continuous, typical ointment structure. However, surprisingly it was found that even up to about 60% the semi-solid systems were composed of definite lipid nanoparticles (Müller et al., 2000a). Similar to aerosil particles, the spherical lipid nanoparticles form a pearl-like network leading to the consistency of cream (Lippacher et al., 2001). The pearl-like structure possesses also the advantage of stabilising the lipid nanoparticle dispersion against aggregation. Lipid nanoparticle dispersions were produced with increasing lipid concentration. The low concentrated dispersions aggregated during storage time, the particles were freely diffusible, collided and led to aggregate

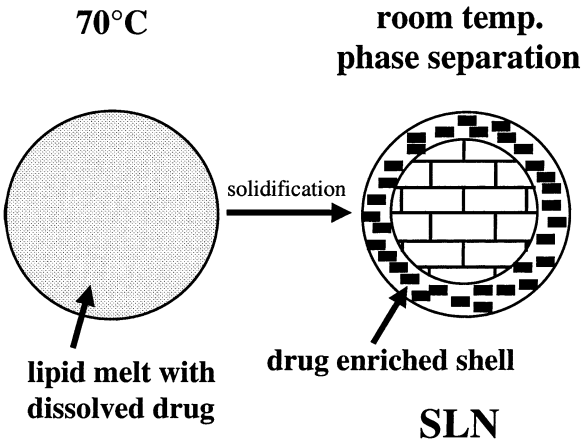

Fig. 4. Phase separation process during cooling in SLN production leading to a drug-enriched shell and consequently burst release.

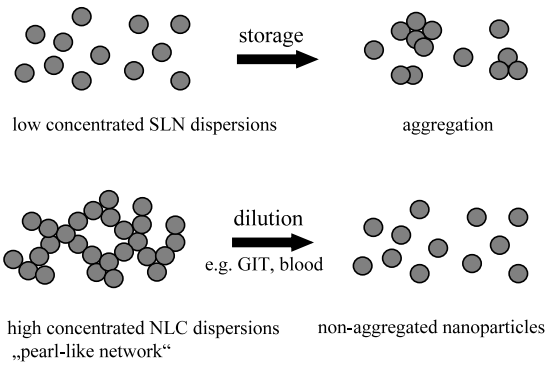

Fig. 5. Aggregation phenomenon in low concentrated SLN dispersions (upper), fixation of high concentrated lipid particle dispersions in pearl-like network (lower), dilution (e.g. in gastric fluid) leads to non-aggregated, definite nanoparticles.

formation (Fig. 5, upper). In the pearl-like network the particles are fixed, diffusion is minimised and thus aggregation avoided. PCS measurements showed no significant increase in particle size after 6 months of storage (Lippacher, 2001).

To further increase the lipid particle concentration, a two-step process was developed. In the first step a high pressure homogenisation is performed, e.g. leading to a particle dispersion of 50% lipid. In a second production step further lipid is dispersed in this 50% dispersion by high speed stirring. Fig. 6 shows the principle of this process starting with a 50% dispersion, e.g. 50 and 50 g water giving a total of 100 g dispersion. In a first addition step 10 g melted lipid are added and dispersed in the 50 g water by stirring, thus leading to 60 g lipid + 50 g water = 110 g dispersion. In the next step 10 g of melted lipid are dispersed in the 50 g water phase leading to 70 g lipid + 50 g water = 120 g dispersion (lipid content now 58%). This process is continued until e.g. 80% lipid concentration is achieved. The total of solid (lipid and solid surfactant) incorporated in the aqueous phase can go up to approximately 95% (Müller et al., 2000a).

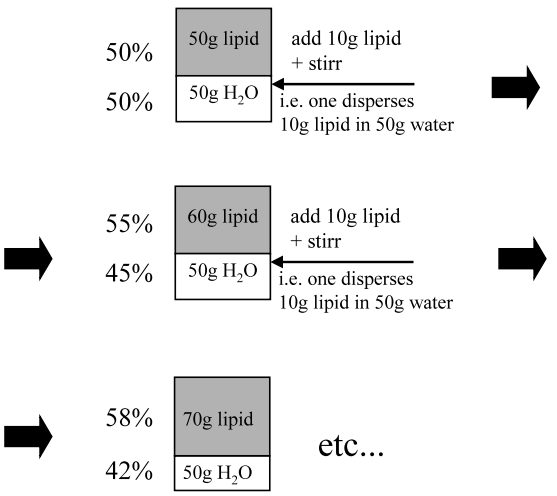

Fig. 6. Two-step production process (homogenisation followed by high speed stirring) to produce highly concentrated lipid nanoparticle dispersion (explanations cf. text).

The highly concentrated particle dispersions have the advantage that distinctly less water needs to be removed when transforming them to e.g. solid dosage forms (e.g. using lipid particle dispersions as granulation fluid or as wetting liquid for the pellet extrusion mass). Basically, this high concentrated process can be applied not only for the third generation particles NLC, but also for the second generation particles SLN.

3.4. Pharmaceutical applications as delivery system

Basically, the NLC can be applied for all applications described for SLN (Müller et al., 1995, 1997, 2000). Areas of highest interest are oral and topical delivery, that means regarding short time-two-market and lowest regulatory hurdles. Full range of excipients is available being of accepted state (e.g. all lipids and surfactants used in creams, tablets, pellets and capsules). Of special interest are life-time extension of drugs (e.g. prednicarbate in topicals, cyclosporine for oral delivery). Due to the special character of NLC, there is no or little possible infringement regarding other patents. There are many patents on emulsions, microemulsions and liposomes, but only a limited number on lipid nanoparticles made from solid lipids, especially made from mixtures of solid and liquid lipids.

Another area of interest is prolonged release of drugs after subcutaneous or intramuscular injection, e.g. erythropoietin. Also dispersions for intravenous injection appear feasible, e.g. using

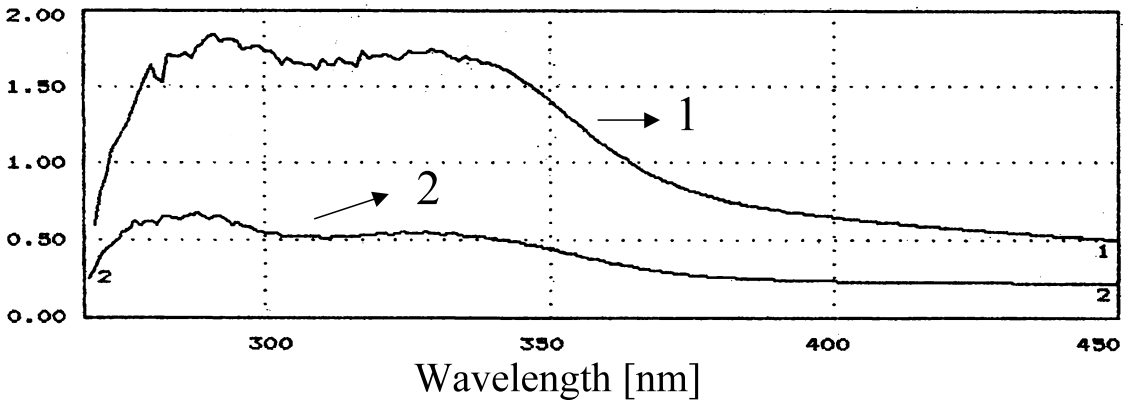

Fig. 7. UV absorption of an O/W emulsion (2) loaded with 1% sunscreen compared with an SLN dispersion (1) loaded with the same amount of sunscreen (after Müller et al., 2000b).

NLC instead of SLN for the formulation of i.v. paclitaxel (Cavalli et al., 2000).

There are also some niche applications, for example ocular delivery of nanoparticles to prolong the retention time. Many papers describe the prolonged retention of drugs in the eye using polymeric nanoparticles, however, up to now no product is on the market due to various reasons (e.g. toxicity problem of non-accepted polymer polyalcylcyanoacrylate). SLN showed an increased retention time in the eye (Gasco, personal communication), it would be even more beneficial to use NLC with improved drug accommodation properties. Similar to nanosuspensions (Jacobs and Müller, 2001) nebulised aqueous NLC dispersions could be exploited for drug delivery to the lungs. With any simple, mechanical nebuliser these dispersions could be administered.

Identical to SLN, a basic advantage of NLC is that a novel encapsulation technology can be combined with a traditional dosage form well known to the patient, e.g. tablet or pellet. The aqueous dispersions can be used as granulation fluid for tablet production or wetting agent for extrusion of pellets.

3.5. Cosmetic applications of NLC

Lipid nanoparticles—SLN and NLC—can be used to formulate active compounds in cosmetics, e.g. prolonged release of perfumes (Müller et al., 2000b; Wissing et al., 2000a). Incorporation of cosmetic compounds and modulation of release is even more flexible when using NLC. In addition, the release of insect repellents has been described (Wissing et al., 2000b; Yaziksiz-Iscan et al., 2002). A feature of general interest is the stabilisation of chemically labile compounds. The solid matrix of the lipid nanoparticle protects them against chemical degradation, e.g. Retinol (Jenning and Gohla, 2001) and coenzyme Q10 (Dingler, 1998).

A recently discovered feature is the sunscreen blocking effect of lipid nanoparticles. Similar to particles such as titanium dioxide the crystalline lipid particles scatter UV light, thus protecting against UV irradiation (Wissing and Müller, 2001). In addition, it was found that incorporation of sunscreens leads to a synergistic UV blocking effect of the particulate blocker lipid nanoparticle and the molecular blocker (Müller et al., 2000b). In vitro, crystalline lipid nanoparticles with the same sunscreen concentration exhibited twice the UV protection effect compared with an O/W emulsion loaded with the sunscreen (Fig. 7).

3.6. Conclusions

The SLN are considered as a versatile nanoparticulate delivery system for different routes of administration, the attractiveness of the system also being documented by the increasing number of research groups world-wide that started work-

ing with this system (Müller et al., 2000). NLC is considered being the smarter, latest generation of lipid nanoparticles possessing improved properties for drug loading, modulation of release profile and stable drug incorporation during storage.

The production technology for highly concentrated lipid particle dispersions—developed in parallel to the NLC technology being applicable to both NLC and SLN—eases the transform of aqueous dispersions to solid products, e.g. tablets, pellets, capsules, but also powders for reconstitution. These advantages are thus overcoming previous existing problems.

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