Introduction

Lipid nanoparticles are intensively investigated as drug delivery systems. However, the presence of supercooled melts and the existence of several polymorphic forms often leads to an unpredictable behaviour of such systems. In addition, many of them are not suitable for heat sterilization. Therefore we investigated the treatment with isostatic (ultra) high pressure as an alternative method for conservation.

Isostatic high pressure treatment (IHT) as a gentle but effective method to decrease microbiological contaminations is a procedure already well known in the food industry [1,2], but also for medical [3] and pharmaceutical purposes. The effects on polymer nanoparticles and thermo sensitive molecules have also been described [4,5] as the treatment of nano suspensions [6].

Our aim was to investigate the behaviour of solid lipid nanoparticles (SLN), a drug delivery system which is in focus since several years, under the influence of isostatic (ultra) high pressure treatment.

Experimental methods

The lipid nanoparticles were produced by melt homogenization in an well known manner [7,8] with a tempered two-stage high pressure homogeniser (Stansted Fluid Power, UK) at 800 bar/100 bar, 20 min, 10°C above the melting temperature of the matrix material.

As matrix materials cetyl palmitate (Cutina CP, donated by Cognis), glycerol behenate (Compritol 888 ATO, donated by Gattefossé S.A.) and hard fat (Witepsol W35 and Witepsol H15, donated by Sasol) were used. Poloxamer 188 (Lutrol F68, donated by BASF AG), Phospholipon 90G and Phospholipon 100H (donated by Phospholipid) were used as surfactants. As a model drug for analytical purposes the lipophilic Epirubicin hydrochloride (EB; Fluka) was incorporated in the nanoparticles.

The obtained colloidal dispersions were divided into 5 ml samples in PP tubes which were high pressure treated in the “Mini Foodlab” FPG620 (SFP, UK), mostly under following conditions: 200 MPa/30 min, 300 MPa/15 min, 600 MPa/8 min, conditions which were already used in [4]. Before and after the HP treatment analyzes with photon correlation spectroscopy (PCS), differential scanning calorimetry (DSC), electron paramagnetic resonance spectroscopy (EPR) were performed.

In contrast to this the system formed with Compritol 888 ATO as matrix is stable even under ultra high pressure conditions, observable in the EPR spectra (Fig. 5). The model drug is partly localized on the particle surface nearby the polar environment but no significant influence of the IHT treatment is observable.

Furthermore cetyl palmitate as matrix material was combined with two phospholipids, Phospholipon 90G and 100H. Both surfactants are suitable for the production of SLN. When treating the SLN with isostatic (ultra) high pressure several differences in the behaviour occurred. The particles prepared with Phospholipon 100H showed a major increase of the particle size caused by the pressure influence. Even rather mild high pressure conditions (200 MPa) lead to a significant increase in size; the polydispersity remained in the same dimension.

Particles prepared with Phospholipon 90G were much more resistant against the high pressure influence. 200 and 300 MPa did not cause significant changes in particle size and polydispersity, whereas a treatment with 600 MPa induced a significant change in particle size. This might indicate a certain instability against very high pressure influence. Temperature effects were excluded in all experiments by setting an upper temperature limit of 50°C during the compression process (automatic stop-start function).

Results and discussion

In a previous work we could show that IHP can cause crystallisation of nanosized supercooled melts [8]. It is therefore an interesting alternative for tedious tempering processes.

Considering the fact that high hydrostatic pressure can have heavy influence on the nanostructure of drug loaded colloidal lipid systems it is in a certain view positively surprising that nevertheless the integrity of the solid particles remains in many cases.

Figure 4 shows moderately resistant system: the marked peak changes slightly depending on the IHT conditions which indicate the model drug to be pushed out more into the polar environment. After 30 min. at 600 MPa a solidification process of the nanodispersion was observed which leads to a particle destruction (see also fig. 6).

Yet another material class for SLN are hard fat matrices. Witepsol W35 was used in a 1:1 mixture with Witepsol H15 and without any mixture to prepare the particles in the known manner with poloxamer 188 as surfactant. Although both materials are suitable for the production of SLN (z-Ave below 200 nm, low polydispersity) distinct differences are observable after the high pressure treatment. While the particles size of W35 particles simply increases slightly under the influence of the (ultra) high pressure treatment the mixture of W35 and H15 does not withstand even moderate pressure conditions (fig. 7).

The results of the various materials show substantial differences in the pressure stress resistance of the particle materials in the chosen combinations with the surfactants. However the stability of the non resistant combinations potentially could be improved by variation of the composition. This is actually under investigation.

Conclusion

Isostatic high pressure treatment as a processing tool for the formulation of lipid nano-dispersions offers several chances. On the one hand it permits the control of crystallisation processes in super cooled systems. On the other hand the decrease of microbiological load for thermolabile nanoparticles is a promising application already often used. However, in contrast to PLGA nanoparticles not all lipid nanoparticle matrix materials withstand the high pressure influence without any qualification. Special investigations for each matrix/surfactant combination are indispensable.

References