Ultrasound preparation of antipsychotic-loaded Solid Lipid Nanoparticles (SLN) for oral delivery

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INTRODUCTION

Some well known antipsychotic drugs which belong to the class II of the biopharmaceutical classification system (Yu, Amidon et al. 2002) have poor water solubility, and thus low absorption trough the gastrointestinal tract. However, these drugs can be formulated by means of oral delivery systems, if taking account on the formulation design in order to ensure consistent bioavailability (Pouton 2006). One strategy to improve oral bioavailability of such drugs would be formulating it by means of lipid-based drug delivery systems, such as solid lipid nanoparticles (SLN), which were developed by Müller and Lucks in the nineties (Müller and Lucks 1996). SLN are derived from o/w emulsions by replacing the liquid lipid (oil) by a solid lipid, i.e., a lipid being solid at both room and body temperatures. These systems represent an alternative to traditional colloidal carriers, such as emulsions, liposomes and polymeric nanoparticles. SLN combine the advantages of the traditional systems, avoiding some of their major disadvantages, e.g., lack of controlled release and long term stability (Müller, Mäder et al. 2000).
The aim of this work was to assess the suitability of SLN, prepared by ultrasound technique, for oral delivery of an antipsychotic drug.

MATERIALS AND METHODS

Imwitor® 900K (glycerylmonoestearate, 40-55%) and Tagat® S (PEG fatty acid esters) were gifts from Sasol (Germany) and Goldschmidt (Germany), respectively. Sodium deoxycholate was purchased from Sigma-Aldrich (Portugal) and the antipsychotic drug was kindly provided from Janssen-Cilag (Belgium). The water used in all experiments was purified, obtained from a MilliQ Plus, Millipore.

Lipid screening tests

The lipid solubility of 1, 2 and 3% (w/w) of antipsychotic drug was evaluated empirically, with 14 different lipids. The most suitable lipid was further analysed by differential scanning calorimetry (DSC), in order to confirm if the drug is dissolved in the lipid phase.
DSC measurements were performed using a Mettler DSC 823e (Mettler Toledo, Switzerland). A sufficient amount of sample was accurately weighted in 40 µl aluminium pan and cold sealed. The reference pan remained empty. Heating curves for the bulk drug and the mixtures of drug and lipid were recorded with a scan rate of 5 K/min from 25ºC to 200ºC and cooled from 200ºC to 25ºC, under liquid nitrogen. For the bulk lipid, heating curves were recorded from 25°C until 85°C at the same increment rate.

Preparation of SLN

SLN were produced containing 10% (w/w) Imwitor® 900K, stabilized with 2.5% (w/w) Tagat S® and 0.5% (w/w) Sodium deoxycholate, by ultrasound technique. Firstly, the solid lipid was heated 5-10ºC above its melting point, and then added to a mixture of surfactants and water, previously heated at the same temperature. A pre-emulsion was obtained under stirring with an Ultra-Turrax

T25 (Janke & Kunkel GmbH, Germany), at 8000 rpm for 5 minutes. This emulsion was further putted under a sonication probe, by means of an Ultrasonic processor VCX130 (Sonics, Switzerland), applying an amplitude of 70% during 20 minutes. For drug-loaded SLN, the drug was added to the solid lipid before melting and sonication. The drug was used in a concentration of 1, 2 and 3% (w/w) with regard to the solid lipid matrix.

Particle size analysis and zeta potential measurements

SLN dispersions were previously diluted with purified water to suitable concentration, and the particle size analysis were performed by photon correlation spectroscopy (PCS), using a Zetasizer Nano ZS (Malvern Instruments, Malvern, UK).
For zeta potential (ZP), the dispersions were diluted with purified water with a conductivity adjusted to 50 µS/cm by dropwise addition of a 0,9% NaCl solution and the zeta potential was accessed by laser Doppler electrophoresis (LDE) with a Zetasizer Nano ZS (Malvern Instruments, Malvern, UK).

RESULTS and DISCUSSION

From the results of lipid screening (data not shown) we conclude that Imwitor® 900K is the most suitable lipid for the preparation of drug-loaded SLN, until a concentration of 3% (w/w), which is the maximum concentration for drug solubility in the lipid (empirically observed). Additionally, no drug crystals were detected in the solidified melts. These results were confirmed by DSC analysis performed in the bulk drug, bulk lipid and in the melted mixtures of lipid with 1, 2 and 3% (w/w) of drug (Table 1).

	Melting point (ºC)	Onset (ºC)	Enthalpy (J/g)
Drug	171.70	170.41	110.81
Inwitor® 900k	63.33	57.05	161.88
Inwitor® 900k + 1% (w/w) drug	61.02	59.18	101.31
Inwitor® 900k + 2% (w/w) drug	60.38	58.05	124.95
Inwitor® 900k + 3% (w/w) drug	60.37	58.22	109.76

Table 1: DSC results of bulk drug, bulk lipid and mixtures of drug with lipid.

The DSC results (Table 1) for the mixtures revealed only the melting events of the lipid, which means that the drug is dissolved in the lipid.

SLN production is normally performed approximately 5°C above the melting point of the lipid (Müller, Mehnert et al. 1995), which means that for the selected lipid, the production temperature was in the range of 75-90 °C. According to DSC results, the melting point of drug is 171.70°C, which means that the existence of drug degradation during the production could be excluded.

Placebo SLN formulations and 1, 2 and 3% (w/w) drug-loaded SLN were prepared, containing 10% (w/w) of lipid phase. The composition of the developed formulations is shown in Table 2.

Formulation [(w/w%)]				
Composition	**DF**	**DL_1**	**DL_2**	**DL_3**
Imwitor® 900K	10.0	9.0	8.0	7.0
Tagat® S	2.5	2.5	2.5	2.5
Sodium deoxycholate	0.5	0.5	0.5	0.5
Drug	-	1.0	2.0	3.0
Water	87.0	87.0	87.0	87.0

Table 2: Composition of prepared SLN formulations (DF, Drug-free SLN dispersions; DL_1, 1% Drug-loaded SLN dispersions; DL_2, 2% Drug-loaded SLN dispersions DL_3, 3% Drug-loaded SLN dispersions).

Particle size analysis (PCS) of prepared SLN dispersions was performed on the production day. Figure 1 shows the PCS diameters (Z-ave) and PI (polydispersity index) of DF, DL_1, DL_2 and DL_3.

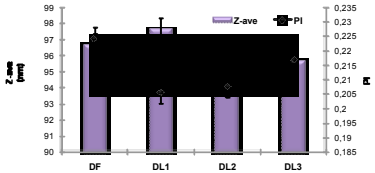

Figure 1: PCS diameters (Z-ave) and PI (polydispersity index) of DF, DL_1, DL_2 and DL_3 formulations, measured on the production day.

Concerning electrochemical stability of dispersions, zeta potential (ZP) of placebo and 1, 2 and 3% (w/w) drug-loaded SLN was evaluated on the production day. Table 3 shows the results of ZP measurements for DF, DL_1, DL_2 and DL_3, formulations, obtained on the production day.

	DF	**DL_1**	**DL_2**	**DL_3**
ZP (mV)	-32.1	-36.0	-33.2	-37.2

Table 3: ZP values of DF, DL_1, DL_2 and DL_3 formulations, measured on the production day.

As we can see from the results, all dispersions have particles in the nanometer range with low PI. The absolute ZP values are relatively high, which predicts a good long-term stability.

CONCLUSIONS

These results show that SLN are suitable carrier systems for the incorporation of poor water soluble antipsychotic drugs, intended for oral administration. The long-term stability studies are going on, in order to determine particles sizes and electrochemical stability of placebo and drug-loaded formulations for longer periods. Additional investigations are also being performed to assess the encapsulation parameters (i.e. encapsulation efficiency and loading capacity), drug release profile from SLN, for further *in vitro/in vivo* correlations studies.

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