Development of nanostructured lipid carriers for therapeutic protein encapsulation and stabilization

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ABSTRACT

Lipid nanocarriers present several advantages compared to other types of nanocarriers, namely, their biocompatibility, high loading capacity and bioavailability, self-assembly and easy production. The lipid nanoparticles developed so far are solid lipid nanoparticles (SLN) and nanostructured lipid carriers (NLC) [1]. The main difference between these two types of nanoparticles is their lipid matrix composition. SLN are exclusively constituted by solid lipid, whereas NLC are constituted by a mixture of solid and liquid lipids. This mixture confers to NLC various advantages when compared to SLN, specifically, higher drug loading, better controlled drug release profile, and minimum unwanted leakage during the nanoparticles shelf-life, due to the resulting imperfect lipid matrix originated by the mixture of solid and liquid lipids. There are numerous benefits on encapsulating therapeutic proteins in nanocarriers, such as protein's protection, enabling specific-target therapies and controlled drug's release [2]. However, little is known about the structure stability of the loaded protein into lipid nanocarriers. The aim of this work was to produce an optimal NLC formulation for protein loading, using a non-heating production method. Insulin was chosen as the therapeutic protein model. The lipid nanoparticles were produced using a modified solvent emulsification−evaporation method based on a w/o/w double emulsion technique[3]. Several combinations of solid lipids, liquid lipids, and surfactants were tested and it was concluded that the best formulation for insulin loaded NLC, with this method, were Suppocire as the solid lipid, and Oleic Acid, as the liquid lipid, in a ratio of 70:30 %, respectively; and Pluronic 1% (w/v) as the surfactant, due to their low mean particle size, 274.5 ± 6.8 nm, a low polydispersity index (PDI) of 0,299 ± 0,0222, indicating particle size conformity, and a zeta potential of -20.1± 7.1 mV, indicating NLC's stability. Furthermore, resorting to HPLC, a high association efficiency (AC) of 70.46 ± 0.27 % and loading capacity (LC) of 2.47 ± 0.01 % was achieved, demonstrating that most of the insulin was incorporated into the NLC. Further studies are being carried out, namely, lyophilization studies with different cryoprotectants, and more importantly protein's structural stability studies.

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