

Role of Solvents and Stabilizer in the Preparation of Solid Lipid Nanoparticles of Stearic Acid by Ultrasound Assisted Nanoprecipitation Method

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ABSTRACT: This article describes the preparation and characterization of solid lipid nanoparticles (SLN) of stearic acid with two different stabilizer systems and mixture of solvents in order to study the influence of solvents and surfactants on the size of the nanoparticles, degree of dispersion, stability and crystallinity of the SLN. A 1:1 mixture of TWEEN-80 and SDS and another stabilizer system containing 1:1 mixture of CTAB and PVP were used as stabilizers. Acetone, mixture of acetone and THF and ethanol were used as solvents. The SLN were characterized by time-resolved particle size analysis, zeta potential measurements, differential scanning calorimetry (DSC) X-ray diffraction (XRD), Transmission electron microscopy(TEM) and FTIR. Results indicate that the SLN produced with the stabilizer system of TWEEN-80 and SDS have lower crystallinity, larger particle sizes, lower melting point as compared with SLN prepared with PVP and CTAB. However the polydispersity indexes for the suspensions are lower than the SLN prepared with PVP and CTAB indicating higher stability. FTIR spectra reveals that there is no structural change occur in all the SLN

KEY WORDS: Solid lipid nanoparticles (SLN), Dynamic light scattering(DLS), stabilizer, TWEEN-80, Transmission electron microscopy(TEM)

I. INTRODUCTION

Solid Lipid nanoparticles (SLN) were developed by Muller in the 1990's. A clear advantage of solid lipid nanoparticles (SLN) over the other colloidal delivery systems such as liposomes, micro emulsions and polymeric nanoparticles [1-3] is the fact that the lipid matrix is made from physiological lipids, which decrease the danger of acute and chronic toxicity [4]. Due to their numerous advantages over existing conventional formulations, SLN have been introduced as a novel carrier system for the controlled release of pharmaceutical and cosmetic actives [5,6]. The occlusive character of solid lipid nanoparticles is another advantage which is due to film formation after application to the skin leading to decreased water of evaporation. Occlusive effect of solid lipid nanoparticles is controlled by particle size, sample volume, lipid concentration and crystallinity of the lipid matrix [7,8]. SLN were also established as an UV radiation protection system due to their particulate character [9]. Incorporation of molecular sunscreens into the matrix of the particles leads to a synergistic effect of molecular sunscreen and the UV light scattering. Thus SLN have potentially lucrative application in topical cosmetic products [10] and as potential drug delivery carrier [2]. Solid lipid nanoparticles of stearic acid [11] finds use as a potential drug carrier [12-13]. The fact that the melting point of this acid is well above human body temperature tends to prevent cosmetics from running and streaking when applied for extended periods.

All classes of stabilizers (with respect to charge and molecular weight) have been used to stabilize the lipid dispersion. It has been found that a combination of stabilizers might prevent particle agglomeration more efficiently [5]. In view of the importance of SLN of stearic acid in preserving moisture of the skin, drug delivery and cosmetic actives carrier, producing stable stearic acid nanoparticles with good crystallinity and stability is significant.

II. EXPERIMENTAL METHODS

Materials

Stearic acid, acetone, tetrahydrofuran (THF), acetonitrile, ethanol were purchased from Merck. TWEEN -80, SDS and CTAB were obtained from Sigma Aldrich. All samples are of analytical grade and hence used as such. Deionized water was used in all preparations.

Method

The SLN's of stearic acid were prepared by a modified nanoprecipitation method. Six experiments were performed. Three different combinations of solvents were used with two stabilizer systems. (I) mixture of CTAB and 0.3% solution of PVP (Mw 40 k Da) and (II) mixture of polyoxyethylene sorbitan fatty acid ester (TWEEN-80) and anionic surfactant sodium lauryl sulphate (SDS). The modified method is defined as follows: On the one hand 125 mg of stearic acid is dissolved in a mixture of solvents (20 mL) (Table .1). On the other hand specified amount of the emulsifier is dissolved (25 ml) in deionized water. The organic phase is added to the aqueous phase with magnetic stirring (200 rpm) at room temperature. Parameters like temperature, stirring speed, rate of addition of organic phase to the aqueous phase are kept constant. The mixture immediately becomes opalescent and was sonicated for 3 minutes (Pci ultrasonic bath sonicator 1.5L). The organic solvents are removed under reduced pressure and the aqueous suspension is centrifuged at 12,000 rpm in a cooling centrifuge (REMI). The obtained precipitate was washed repeatedly with water and it was re-dispersed in deionized water by sonication and the suspension was stored in vials. The preparation parameters (solvent compositions and stabilizers) of the six batches of stearic acid nanoparticles their characterization and physicochemical studies, are presented in Table- 1

III. RESULTS AND DISCUSSION

Effect of stabilizer and solvent on the size, morphology and crystallinity of the SLN

According to TEM images, (Tecnai-12 G2 with EDAX Bio Twin TEM from FEI Company tools for Nanotech.) the particles have a smooth surface and are spherical, and the majority of the particles do not exhibit aggregation (Figures. 1 (a)-(f)). All the six formulations of SA SLN were crystalline as shown from the XRD (RICHSEIFER) pattern which is in agreement with the literature values of FCC crystal structure [JCPDS Card No. 09-0618] of stearic acid (Figure.2). There are no additional peaks indicating absence of other lipid modifications. The average particle size was found to vary from 100 nm (SA-1) to 300 nm (SA-6) as determined by dynamic light scattering (DLS) (Malvern Zetasizer nano -S). Smaller particles are formed in the presence of stabilizer system-I in the same solvents. Wolfgang *et al.* [14] reported cationic stabilizers producing smaller particles. However, the poly dispersity index (P.I) of the SLN is lower with stabilizer system II (TWEEN-80 and SDS) (SA-1-0.772, SA-2-0.773, SA-3-0.399, SA-4-0.595, SA-5-0.404, SA-6-0.350) indicating higher stability.

Thermograms of the six SA SLN are given in Figure. 3. The physical state of the dispersed particles of the SLN are understood via the characteristic melting or transformation endotherms upon heating.

Table - 1
Preparation parameters and physico chemical characteristics of the stearic acid (SA) nanoparticles

Parameters	SA-1	SA-2	SA-3	SA-4	SA-5	SA-6
Water	25mL	25mL	25mL	25mL	25mL	25mL
Acetone	18mL			18mL		
Ethanol		20mL			20mL	
Acetonitrile			15mL			15mL
Tetrahydrofuran (THF)	2mL		5mL	2mL		5mL
CTAB	2mL	2mL	2mL			
PVP	2mL	2mL	2mL			
TWEEN-80				0.5mL	0.5mL	0.5mL
SDS				0.3mL	0.3mL	0.3mL
Mean particle size obtained from DLS Z_{ave} (nm)	271.6	205.1	355.8	320.0	323.5	323.5
Particle size obtained from TEM, (nm)	100	150-180	200	200	250-280	280-300
M.pt from DSC measurement	61.1°C	64.2	60.9°C	54.1°C	59.5°C	52.4
Enthalpy of formation J/g	181.3	86.17	140.9	49.75	70.82	35.75
Zetapotential(mV)	-18.3	-12.6	+29.0	-13.0	-16	-8.3

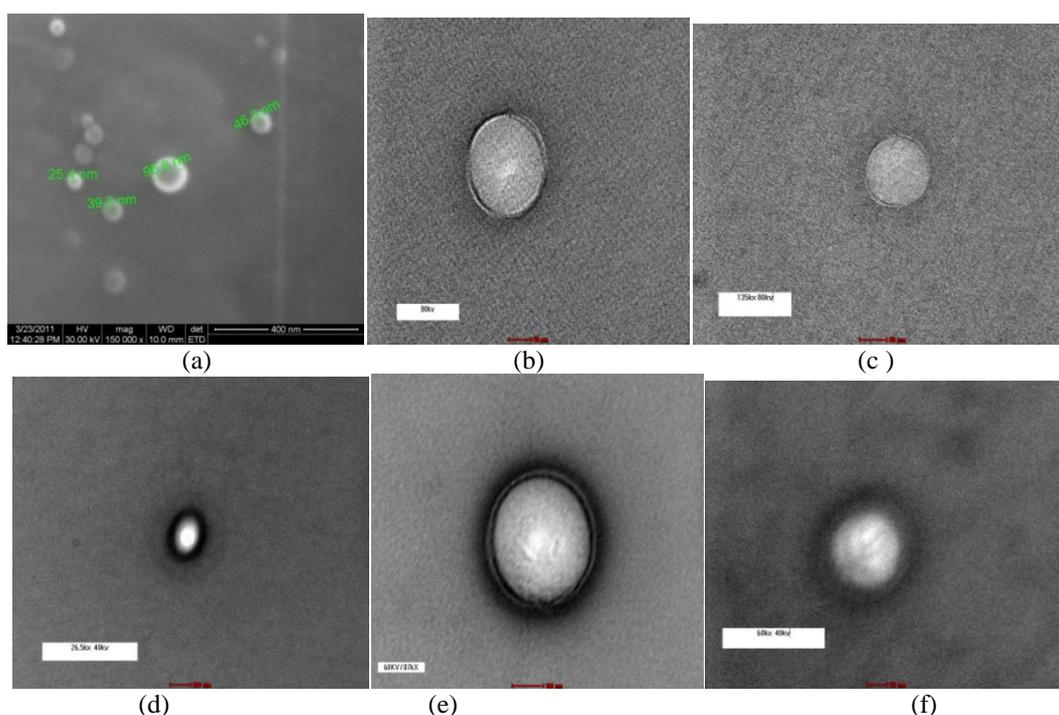


Fig.1 SEM images of SLN (a) SA-1 of average size 100 (scalbar 400 nm) TEM images of (b) SA-2, 150-180 nm (scale bar 50nm) (c) Nanosphere of SA-3 size 200-250nm (scale bar 50nm) (d) SA-4, 200nm (scale bar 200nm) (e) A single nanosphere of SA-5 250-280 nm (f) Nanosphere of size of SA-6 280-300nm (scale bar 100 nm)

The DSC (NETZSCH DSC204) measurements indicate that the melting point of the stearic acid particles prepared with TWEEN-80 and SDS (52.4-59.5 °C) are lower than that of the nanoparticles prepared with PVP and CTAB (60.9 -64.2 °C). Thus it is clear that the stabilizer system-II (TWEEN-80 and SDS) reduces the crystallinity and melting point of the SA SLN formulations. Presence of a single peak indicates the absence of other lipid modifications. The zeta potential values ranging from -8.3 mV and +29mV indicates moderate stability of the SA SLN with limited flocculation [15]. The irreversible transition of stearic acid at about 80°C was not observed in the particle form of stearic acid prepared with the above mentioned stabilizer blends.

The FTIR (Perkin Elmer) spectra of the six SA SLN are given in Figure.4. All the characteristic absorption bands of stearic acid are seen in the FTIR spectra six SLNs. The broad

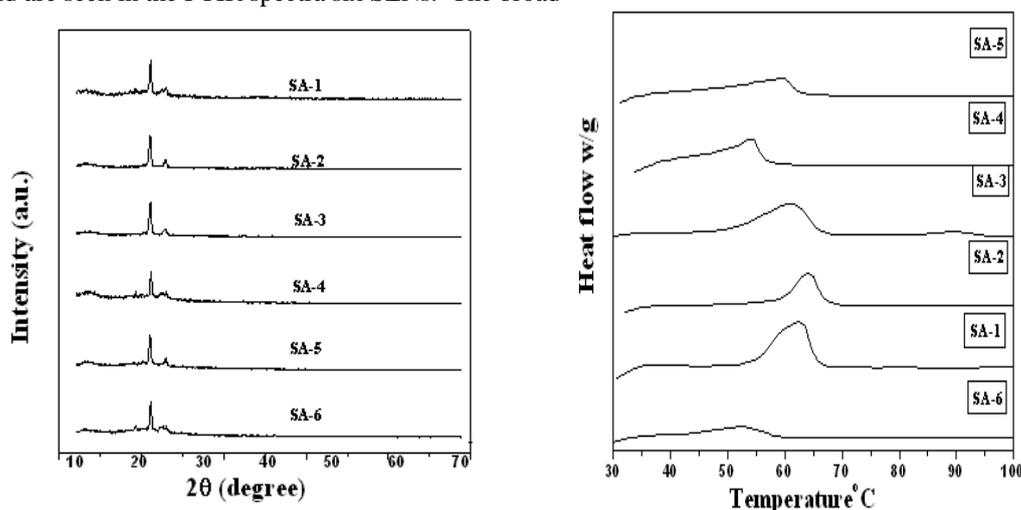


Fig. 2 PXRD of the six SLN of SA-1 to SA-6 Fig. 3 Thermograms of the six SLN SA-1 to SA-6

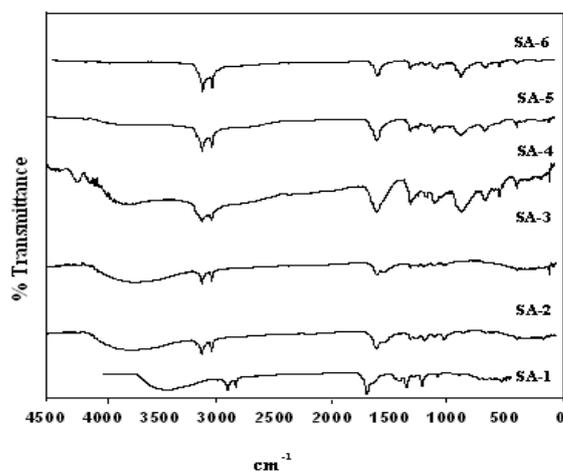


Fig. 4 FTIR spectra of the SLN of stearic acid SA-1 to SA-6

band owing to the O-H_{str} at 3000-3300 cm⁻¹ is significant in the first four formulations with slight variations but it is shifted to higher frequency and not well defined in the formulations SA-5 & SA-6. This may be attributed to the formation of nanoparticles.

IV. CONCLUSION

Six formulations of Stearic acid nanoparticle suspensions (SA-1 to SA-6) were prepared with chosen mixture of solvents and a combination of stabilizers. Among the chosen stabilizers, for a specific mixture of solvents the SLN prepared with CTAB and PVP is found to be smaller in size, more crystalline and has higher melting point in comparison with the SLN prepared with TWEEN-80 and SDS as emulsifier. However polydispersity index measurements show that SLN prepared with TWEEN-80 and SDS are more stable and hence have long storage time. Among the chosen solvents for a given stabilizer, mixture of acetone and THF in the ratio 9:1 produced smaller particles with good crystallinity. Further work involves incorporation of the SLN in creams and *invitro* study for skin hydration.

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