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FORMULATION AND EVALUATION OF SOLID NANO EMULSION OF FUROSEMIDE

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ABSTRACT

The aim of the present work is to prepare solid nanoemulsion of a poorly water soluble drug Furosemide with aerosil as carrier. Based on solubility studies and pseudo ternary phase diagrams, five liquid SNEDDS were prepared with selected systems in various proportions and evaluated for self emulsification time, phase separation and precipitation of the drug, robustness to dilution, percentage transmittance, thermodynamic stability studies, droplet size, PDI and zeta potential. From the evaluation studies it was found that formulation consisting of oleic acid(15.4%w/w), tween80(57.1%w/w), PEG400 (17.5%w/w) and drug(10%w/w) was stable and optimum and selected for preparation of S-SNEDDS. With selected optimum formulation solid nanoemulsion is prepared using aerosil as carrier in 1:2 ratio by adsorption technique and evaluated for flow properties, drug content, effect of dilution, droplet size determination, FT–IR studies, in–vitro drug release study and accelerated stability study for 6 months. Prepared solid nanoemulsion showed "good" flow properties and 95.162±1.24% drug content. Reconstitution properties showed spontaneous nano emulsification with droplet size 65.02 nm and PDI 0.648. Results of in-vitro dissolution revealed that % drug release from solid nanoemulsion was higher than that of pure drug and marketed tablet. Results of accelerated stability study for 6 months showed that formulation was stable and no alteration in the dissolution rate was observed. The results of present study have proved the potential

use of solid nanoemulsion to improve solubility and dissolution rate of poorly water soluble drug furosemide.

KEYWORDS: Solid nanoemulsion, Aerosil, Dissolution rate, accelerated stability Study and Furosemide.

1. INTRODUCTION

Therapeutic effectiveness of drug depends on bioavailability and ultimately upon the solubility of drug molecules. Solubility is one of the important parameter to achieve desired concentration of drug in systemic circulation for pharmacological response to be shown. Currently only 8% of new drug candidates have both high solubility and permeability.[1] Various approaches should use to improve the dissolution rate of the drug. Among them, Self nano emulsifying drug delivery systems (SNEDDS) have shown great pledge for enhancing bioavailability of poorly soluble compounds.[2-3] SNEDDS[4-7] are isotropic and thermodynamically stable solutions consisting of an oil[8], surfactant, co-surfactant and drug mixtures that spontaneously forms oil in- water nano emulsions[9] when mixed with water under gentle stirring. The basic principle of this system is its ability to form fine oil-in-water (o/w) nano emulsion under gentle agitation following dilution by aqueous phases. This spontaneous formation of an emulsion in the GI tract presents the drug in a solubilized form, and the small size of the formed droplet provides a large interfacial surface area for drug absorption. Particularly for BCS class II substances, the bioavailability may be enhanced by increasing the solubility and dissolution rate of the drug in the gastro-intestinal fluids. SNEDDS are generally encapsulated either in hard or soft gelatin capsules. SNEDDS may interact with the capsule resulting in either brittleness or softness of the shell.[10] To overcome this problem SNEDDS need to convert into Solid SNEDDS. Many techniques are offered to convert conventional liquid SNEDDS to solid such as adsorption to solid carriers, spray drying, spray cooling, melt granulation, rotary evaporation, freeze drying and high pressure homogenization.[11-14] But adsorption process is simple and involves simply addition of the liquid formulation to solid carriers by mixing in a blender. The resulting powder may then be filled directly into capsules or mixed with suitable excipients before compression into tablets.

Furosemide is a potent diuretic agent that induces a powerful diuresis, followed by the loss of sodium, potassium and chloride into the urine by acting on thick ascending limb of the loop of henle. It is used in the treatment of both acute and chronic renal failure. Its variable oral absorption is about 11-60% due to insufficient aqueous solubility at gastro intestinal pH.

Therefore solid nanoemulsions serves as a tool for the delivery of poorly aqueous soluble drug furosemide, without affecting invitro drug release compared to that of liquid nanoemulsion.

The aim of the present study was to develop novel solid nano emulsion from liquid nanoemulsion formulation containing furosemide. The objective was to design a solid nanoemulsion of furosemide that could improve its dissolution rate and stability.

2. MATERIALS AND METHODS

2.1. Materials

Furosemide, Cremophere RH 40, cotton seed oil, soya bean oil, aerosil and meglyol were obtained as gift samples from Bright Labs pvt. Ltd. (Hyderabad, India). Tween 20, Tween 80, PEG 400 and oleic acid were purchased from Merck specialities Pvt. Ltd. (Mumbai, India.) Span 80, propylene glycol and potassium dihydrogen phosphate were purchased from S.D Fine Chemicals (Mumbai, India.).

2.2. Methods

2.2.1. Solubility Studies

The saturation solubility of furosemide was determined in selected vehicles such as oils, surfactants and co- surfactants. 1g of vehicle was taken in a screw capped glass vial and to this excess amount of drug was added. The resultant drug-vehicle mixtures were cyclomixed using cyclomixer (CM 101 DX) to aid solubilization. The mixtures were then heated in a thermostatic water bath at 40°C for 10 minutes to facilitate solubilization. Then the mixtures were shaken by using orbital shaking incubator (VIGNAN-OSR30) at 25°C for 48hrs and kept for equilibration for another 24 hrs. After reaching equilibrium each vial was centrifuged at a speed of 3500 rpm for 15 minutes. The supernatant was separated by filtration using 0.45µ filters and then suitably diluted with methanol. Samples were analyzed spectrophotometrically[15,16] at 273 nm. Concentration of furosemide in each vehicle was calculated using regression equation that was given along with previously constructed calibration curve.

2.2.2. Construction of Pseudo-Ternary Phase Diagram

Pseudo- ternary phase diagrams were constructed for selected oil, surfactant, co-surfactant with water at room temperature by water titration method. From solubility studies oleic acid was selected as oil phase; Tween 80, PEG400 were selected as surfactant and co-surfactant

respectively. The surfactant was mixed with co- surfactant in the ratio of 4:1, 3:1, 2:1 and 1:1 respectively. Aliquots of surfactant/co-surfactant mixture was then mixed with oil at ratios of 9:1, 8:2,7:3, 6:4, 5:5, 4:6, 3:7, 2:8 and 1:9 in different vials and then titrated with water at room temperature.[17] The samples were then equilibrated for 30 seconds and visually observed after each addition. Based on visual observation the systems were classified as nanoemulsion, micro emulsion, coarse dispersion and gel formulation.[18] Pseudo ternary phase diagrams were then constructed using Triplot software version 4.1.2. The samples which were clear or bluish transparent in appearance were considered as nanoemulsions.

2.2.3. Preparation of liquid SNEDDS

Five different SNEDDS of furosemide were prepared using oleic acid as oil, Tween 80 as surfactant and PEG 400 as co-surfactant. Concentration of Furosemide was kept constant (10mg) in all formulations. Surfactant/co-surfactant mixture (Smix) was prepared by mixing in suitable proportions and cyclomixed. Furosemide was accurately weighed and dissolved in oil and then Smix was added to above oil-drug mixture. The components were cyclomixed until transparent preparations were obtained. Finally prepared SNEDDS of Furosemide were kept aside at room temperature to examine for signs of turbidity (or) phase separation prior to characterization.

2.3. Characterization of liquid SNEDDS

2.3.1. Self emulsification and visual assessment

Self emulsification property of SNEDD formulations[19] was evaluated by visual assessment. Time taken for the formation of nano emulsion was determined by drop wise addition of formulation to 250 mL of distilled water, simulated gastric fluid and phosphate buffer of pH 6.8 in separate glass beakers at 37°C and the contents were gently stirred using magnetic stirrer at 100rpm. The tendency to form an emulsion is assessed as “good” when emulsification occurs rapidly in less than 1 minute with clear (or) transparent appearance. The tendency to form an emulsion is assessed as “bad” when there is less clear emulsion formation. Depending on visual appearance and time taken for self emulsification, formulations are graded as.[20]

Grade A: Rapidly forming (within 1min) nano emulsion having a clear (or) bluish appearance; Grade B: Rapidly forming, slightly less clear emulsion, having a bluish white appearance; Grade C: Fine milky emulsion that formed within 2 minutes; Grade D: Dull, grayish white emulsion with a slight oily appearance that is slow to emulsify (more than 2

minutes); Grade E: Formulation Exhibiting either poor or minimal emulsification with large oil globules present on the surface.

2.3.2. Phase separation and stability study of emulsions

Each SNEDDS formulation (50µL) was added to a vials containing 5mL of double distilled water, simulated gastric fluid at room temperature and cyclomixed for 1 minute then each mixture was stored and observed for phase separation and precipitation of drug at intervals 2, 4, 6, 8, 12, 24 h period of time.

2.3.3. Robustness to dilution

Prepared SNEDDS formulations were subjected to dilution[21] in ratios 1:100 and 1:1000 folds with distilled water, 0.1 N HCl and phosphate buffer of pH 6.8. The diluted nano emulsions were stored for 24 h and visually observed for any signs of phase separation (or) precipitation of drug.

2.3.4. Drug loading efficiency

Drug content in formulation was determined UV Spectrophotometer. 50 mg of each formulation was accurately weighed and dilute to 100mL with methanol. Resultant solutions were analyzed spectroscopically after suitable dilution. Drug loading efficiency[22] was calculated by equation

$$\text{Drug loading efficiency} = \frac{\text{Amount of drug in known amount of formulation}}{\text{Initial drug load}} \times 100 \quad \textbf{(1)}$$

2.3.5. Thermodynamic stability studies

The prepared SNEDDS formulations were subjected to thermodynamic stability studies to study the effect of centrifugation and temperature on stability of nano emulsions.

2.3.6. Centrifugation study

The formulations were added to deionized water in the ratio 1:20 and centrifuged at 3500 rpm for 30 minute and observed for phase separation (or) precipitation.[23]

2.3.7. Freeze thaw cycle

The formulations which are stable under centrifugation were subjected to freeze thaw cycle.[24] In this study, SNEDDS formulations were diluted with deionized water in 1:20 ratio and subjected to two freeze thaw cycles between -20°C and +25°C by storing at each

temperature for 48hrs and after 48hrs samples were observed for phase separation (or) precipitation.

2.3.8. Droplet size and Zeta potential determination

Prepared SNEDDS formulations were added to distilled water in ratio 1:1000 in test tube and mixed for 1 minute using a cyclo mixer. The droplet size, PDI of the emulsions were determined at 25°C by dynamic light scattering (DLS) technique at 90◦ angle and Zeta potential was determined by electrophoretic light scattering technique using Zeta sizer 3000 HAS.

2.4. Formulation of Solid – SNEDDS

From the characterization studies done on five different furosemide SNEDDS[25], the formulation with good stability, good self nano emulsification property with less particle size and less PDI was selected to formulate as solid nanoemulsion. Solid nanoemulsion was prepared by mixing liquid SNEDDS containing furosemide with Aerosil as carrier in ratio of 1:2. Liquid SNEDDS was added in drop wise manner over Aerosil contained in porcelain dish. After each addition, contents were mixed using glass rod for uniform distribution of formulation. Resultant damp mass was passed through sieve # 120 and dried at room temperature and stored until further use.

2.5. Characterization of S-SNEDDS

2.5.1. Flow properties of S-SNEDDS

The angle of repose[26-28] of S-SNEDDS was determined by funnel method. Height of funnel was adjusted in such a way that the tip of the funnel just touches the apex of the heap of powder. Accurately weighed sample was allowed to flow through funnel freely onto the surface. The diameter of the powder cone was measured and angle of repose was calculated using the equation

$$\tan\theta = \frac{h}{r} \qquad \textbf{(2)}$$

where h and r are height and radius of powder cone.

2.5.2. Bulk density and tapped density

A quantity of 2gm of S-SNEDDS was introduced into 10mL measuring cylinder. Initial volume was noted and cylinder was allowed to fall under its own weight into a hard surface from a height of 2.5 cm at 2 second intervals. Tapping was continued until no further change

in volume was noted. Bulk density and Tapped density were calculated using the following equations[22];

$$\text{Bulk density (BD)} = \frac{\text{Weight of powder blend}}{\text{Volume of the packing}} \quad \textbf{(3)}$$

$$\text{Tapped density (TD)} = \frac{\text{Weight of powder blend}}{\text{Tapped Volume of the packing}} \quad \textbf{(4)}$$

2.5.3. Compressibility index

The compressibility index of the blend was determined by Carr's compressibility index given by the equation.

$$\text{Carr's compressibility index (\%)} = \frac{\text{TD - BD}}{\text{TD}} \times 100 \quad \textbf{(5)}$$

Hausner's Ratio Hausner's Ratio is a number that is correlated to the flowability of a powder (or) granular material. Hausner's ratio can be calculated by the equation

$$\text{Hausner's Ratio} = \frac{\text{TD}}{\text{BD}} \quad \textbf{(6)}$$

2.6. Drug content

S-SNEDDS of Furosemide was accurately weighed equivalent to 40mg and dissolved in sufficient quantity of methanol. The solution was sonicated for 10min in order to extract the drug in methanol and filtered.[29] The absorbance of filtrate was measured at 273 nm using UV-Visible Spectrophotometer.

2.7. Reconstitution properties of S-SNEDDS

2.7.1. Effect of dilution on S-SNEDDS

100 mg S-SNEDDS was accurately weighed and introduced into 100 mL double distilled water[29] in a beaker at 37°C and mixed gently using magnetic stirrer at 100rpm. The property of rapid emulsification was observed. The tendency to form an emulsion is assessed as "good" when emulsification occurs rapidly in less than 1 minute with clear (or) transparent appearance. The tendency to form an emulsion is assessed as "bad" when there is less clear emulsion formation.

2.7.2. Droplet size determination

100 mg of S-SNEDDS formulation was diluted with 100 mL distilled water in a test tube and cyclomixed. The droplet size and poly dispersibility index(PDI) of emulsion was determined

at 25°C by dynamic light scattering (DSC) technique at 90◦ angle using a Zeta sizer 3000 HAS.

2.7.3. FT-IR studies

FT-IR Spectrum of pure drug, Aerosil and Formulation were obtained by FT-IR Spectrophotometer. The spectrums were taken with the accumulation 24 scans and a resolution of 4 cm^{-1} over the range of 400-4000 cm^{-1}. The spectrum of formulation so obtained was compared with spectrum of pure drug for any interactions.

2.8. In-Vitro drug release study

The in-vitro dissolution study of S-SNEDDS which were filled into 0 size capsule, API and marketed drug were carried out using USP-Type I dissolution test apparatus (Lab India DS 8000) in 500 mL buffer of pH 1.2 containing 0.3% w/w SLS at 37±0.5°C with 100 rpm. Samples were withdrawn at 5, 10, 15, 30, 45 and 60 min time intervals and filtered through 0.45µm filter. An equal volume of dissolution medium was replenished after every sampling to maintain constant volume. Samples were analyzed using a double beam UV-Spectrophotometer at 273 nm. The cumulative percentage drug release was calculated and graph was plotted against time.

2.9. Accelerated stability Testing

Accelerated stability studies of S-SNEDDS formulations were carried out according to ICH guidelines. The formulation was stored at 40°C and 75% RH for 6 months in stability chamber. Later the formulation was evaluated for parameters such as effect of dilution, droplet size, PDI and in vitro drug release.

3. RESULTS AND DISCUSSION

3.1. Solubility studies

Solubility of Furosemide was determined in various Oils, Surfactants, Co-surfactants by UV-Spectrophotometric method. Furosemide has shown maximum solubility in oils Oleic acid, in surfactant tween 80 and in co-surfactant PEG 400. Results of solubility studies are given in Figure 1 and 2.

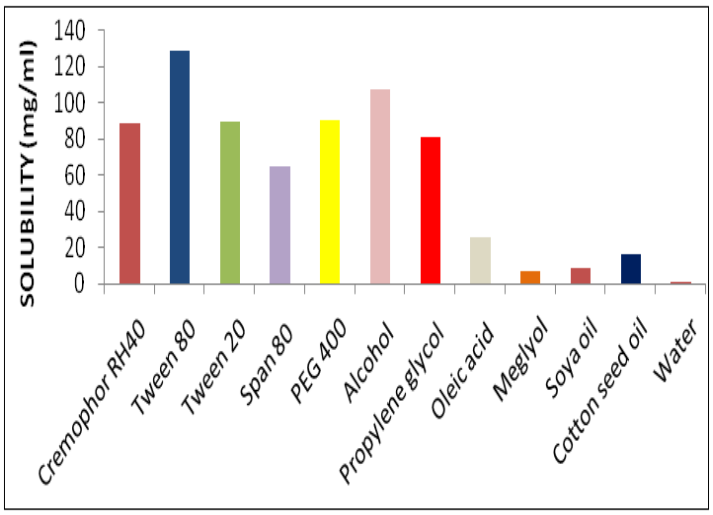

Fig. 1. Solubility of furosemide in various oils

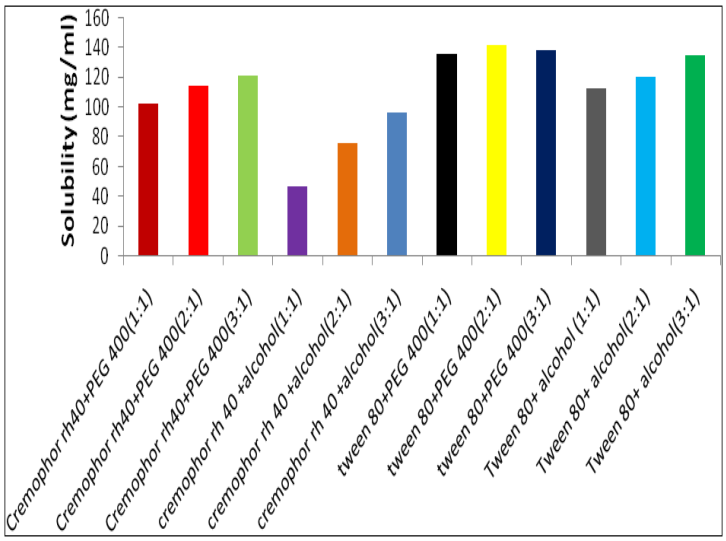

Fig. 2. Solubility of Furosemide in various Surfactants

3.2. Construction of Pseudo-ternary Phase Diagrams

Pseudo–ternary Phase Diagrams were constructed to identify the nano emulsion regions and to identify suitable composition of oil, surfactant and co-surfactant for formulation of SNEDDS. From Pseudo–ternary phase diagrams it has been found that the systems consisting of oleic acid as oily phase, Tween 80 as surfactant and systems PEG400 as co-surfactant showed good nano emulsifying property. It was also found that by increasing oil content systems show the appearance of coarse emulsion. It was also found that for systems consisting of Oleic acid, Tween 80 and PEG 400 the increase in cosurfactant proportion in Smix systems showed decreasing property of spontaneous nano emulsion formation. From this observation it was also clear that Surfactant play role to form nano emulsion in a proper range spontaneously. Pseudo-ternary phase diagrams of NE are shown in **Figure 3.**

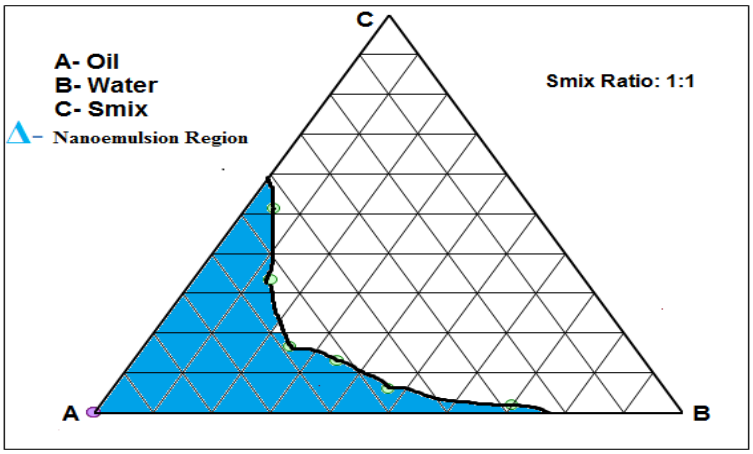

Fig. 3. Pseudo ternary phase diagram of SNE 2.

3.3. Preparation of selected SNEDDS formulations

Five different SNEDDS formulations are prepared with varying ratios of Oil, Surfactant+ Co-surfactant and water. In all formulations the amount of furosemide is constant (40mg). Composition of prepared SNEDDS formulations was given in **Table 1.**

Table 1. Composition of prepared SNEDDS formulations

Formulation Code	Drug (mg)	Oleic Acid (%w/w)	Smix (1:1) (%w/w)	Water (%w/w)
SNE1	40	43.9	4.87	51.28
SNE2	40	53.3	13.3	33.33
SNE3	40	58.3	25	16.6
SNE4	40	52.17	34.7	13.04
SNE5	40	46.72	46.7	6.04

3.4. Evaluation of Furosemide liquid SNEDDS

3.4.1. Self emulsification and visual assessment

According to visual assessment formulations are graded for self-emulsification time. Self emulsifying mixtures should disperse rapidly in aqueous medium with mild shaking. Self emulsification time was determined for prepared SNEDDS and was given in **Table 2**. It was found that all formulations are emulsified in 30 to 42 seconds i.e. performance of all formulations was said to be good.

Table 2. Self emulsification time (Seconds) (n=3)

Formulation Code	Self emulsification time (sec)	Performance of emulsion
SNE1	38.23±0.35	Good
SNE2	35.45±0.28	Good
SNE3	40.87±0.34	Good
SNE4	41.24±0.56	Good
SNE5	41.32±0.87	Good

3.4.2. Phase separation and precipitation study of emulsions

Prepared SNEDDS formulations were observed for precipitation and phase separation of drug at intervals 2, 4, 6, 8, 12, 24 h period of time and it was found that all formulations showed neither precipitation nor phase separation of the drug. Results are given in **Table 3.**

Table 3. Phase separation and precipitation of the drug (n = 3)

Formulation code	Precipitation	Phase Separation
SNE1	No	No
SNE2	No	No
SNE3	No	No
SNE4	No	No
SNE5	No	No

3.4.3. Robustness to Dilution

Formulations are diluted with excess of Water, 0.1N HCl and Phosphate buffer of pH 6.8 and the diluted samples are stored for 24hrs and visually observed for precipitation (or) phase separation of drug. No precipitation (or) phase separation is found which indicates all formulations are robust to dilution. Results are given in Table 4.

Table 4. Robustness to dilution (n = 3)

Formulation Code	Distilled Water	0.1N HCl	Phosphate buffer pH 6.8
SNE1	Pass	Pass	Pass
SNE2	Pass	Pass	Pass
SNE3	Pass	Pass	Pass
SNE4	Pass	Pass	Pass
SNE5	Pass	Pass	Pass

3.4.4. Drug loading efficiency

It was found that all formulations have drug loading efficiency more than 90% & the Results are given in Table 5.

Table 5. Drug loading efficiency of formulations (n=3)

Formulation Code	Drug loading efficiency
SNE1	92.76±0.67
SNE2	98.67±0.98
SNE3	95.45±0.56
SNE4	97.59±0.45
SNE5	93.23±0.23

3.4.5. Thermodynamic stability studies

Thermodynamic stability study is designed to identify metastable formulation. The SNEDDS are subjected to Centrifugation study and Freeze thaw cycle. The emulsions are stable during centrifugation at 3,500rpm and alternative temperature cycles of -20°C and +25°C. There is no precipitation and phase separation of formulations. The results are given in **Table 6.**

Table 6. Thermodynamic stability studies of SNEDDS

Formulation code	Centrifugation (3500rpm for 30min)	Freeze Thaw cycle (-20°C and +25°C)
SNE1	Pass	Pass
SNE2	Pass	Pass
SNE3	Pass	Pass
SNE4	Pass	Pass
SNE5	Pass	Pass

3.4.6. Droplet size and Zeta potential determination

Droplet size, PDI and Zeta potential of the prepared formulations were determined. Droplet size was found to be in between 65 to 96 nm and PDI of all formulations was found to be below 1.0 indicating that the particles are distributed uniformly. Zeta potential was found to be in between – 3.25 to - 10.69 mV. The results are given in **Table 7.** From the results it was found that formulation SNE-2 showed less droplet size than other formulations.

Table 7. Results of Droplet size, PDI and Zeta potential(n=3)

Formulation code	Mean droplet size (nm)	PDI	Zeta potential(mV)
SNE1	70.84	0.942	-6.78
SNE2	65.02	0.648	-3.25
SNE3	68.08	0.871	-10.69
SNE4	96.56	0.728	-8.97
SNE5	87.27	0.812	-4.68

3.4.7. Preparation of Solid SNEDDS of Furosemide

Based on evaluation tests done for five liquid SNEDDS formulations the formulation SNE-2 was selected for preparation of solid SNEDDS of Furosemide. Compared to other formulations SNE-2 showed good self emulsification property which was emulsified spontaneously in 35.45±0.28 sec and also droplet size (65.02 nm) was less than other formulations with more uniform distribution of particles (PDI = 0.648). From the evaluation studies it was found that formulation consisting of oleic acid (15.4%w/w), tween 80(57.1%w/w), PEG400 (17.5%w/w) and drug (10%w/w) was stable and optimum and

selected for preparation of S-SNEDDS. With selected optimum formulation s-SNEDDS are prepared using aerosil as carrier in 1:2 ratio by adsorption technique.

3.5. Evaluation of solid SNEDDS of furosemide

3.5.1. Flow properties of s-SNEDDS

Flow properties such as Angle of Repose, Bulk density, Tapped density, Compressibility Index and Hausner's Ratio are determined and it was found that Prepared s-SNEDDS showed "**Good"** flow properties. Results are given in **Table 8**.

3.5.2. Drug Content

Amount of drug present in prepared solid nanoemulsion was determined. Drug content of the s-SNEDDS was found to be **95.136 ± 1.29**%.

Table 8. Flow properties of solid nanoemulsion of Furosemide (n=3)

FLOW PROPERTIES	RESULTS
Angle of repose(°)	20.10±0.08
Bulk density(g/ml)	0.378±0.016
Tapped density(g/ml)	0.412±0.012
Compressibility index	10.34±0.87
Hausner's ratio	1.12±0.56

3.5.3. Effect of dilution on s-SNEDDS

Effect of dilution on solid nanoemulsion was studied and it was found that prepared solid nanoemulsion showed spontaneous emulsification i.e. in less than 1min and it was also found that there is no phase separation (or) phase inversion of nano emulsion after 24hrs storage of diluted sample.

3.5.4. Droplet size Determination

Mean droplet size and Poly dispersibility index of reconstituted solid nanoemulsion were found to be 64.98nm and 0.646. The solid nanoemulsion showed PDI less than 1.0 i.e. there is distribution of uniform size particles.

3.5.6. FT – IR Studies

FT-IR Spectrum of pure drug, Aerosil and the sSNEDDS were obtained by FT-IR Spectrophotometer. The spectrum of s-SNEDDS so obtained was compared with spectrum of pure drug for any interactions. Characteristic peaks observed **at 1563, 3284, 3351** cm^{-1} sulfonamide group peak and for OH stretching vibration. FT-IR spectrum of pure drug and s-

SNEDDS were almost similar because of same functional groups. It indicates there was no interaction between Furosemide and excipients used in formulation. FT-IR spectrums of pure drug, Aerosil and solid nanoemulsion are shown in Figures 4.

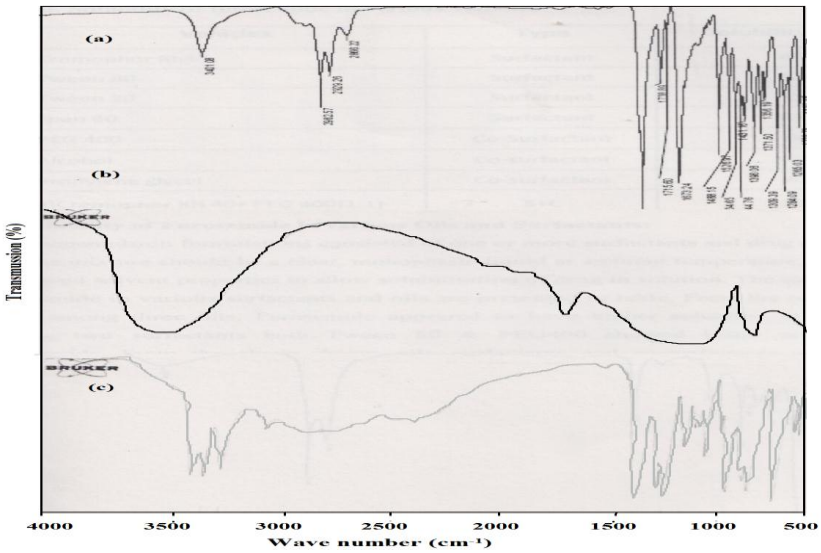

Fig. 4. (a) FT – IR Spectrum of solid nanoemulsion; (b) FT – IR Spectrum of Aerosil; (c) FT-IR Spectrum of pure drug (Furosemide)

3.5.7. In – Vitro drug release study

In – Vitro drug release study was done for pure drug, marketed tablet and s-SNEDDS of Furosemide. The percentage drug release from s-SNEDDS was found to be higher than that of pure drug and marketed tablet. Cumulative Percentage drug release (Fig. 5) from prepared s-SNEDDS at 60 min was found to be 99.51 ± 1.24% whereas for pure drug and marketed tablet it was only 16.28 ± 2.54% and 64.72 ± 2.06%.

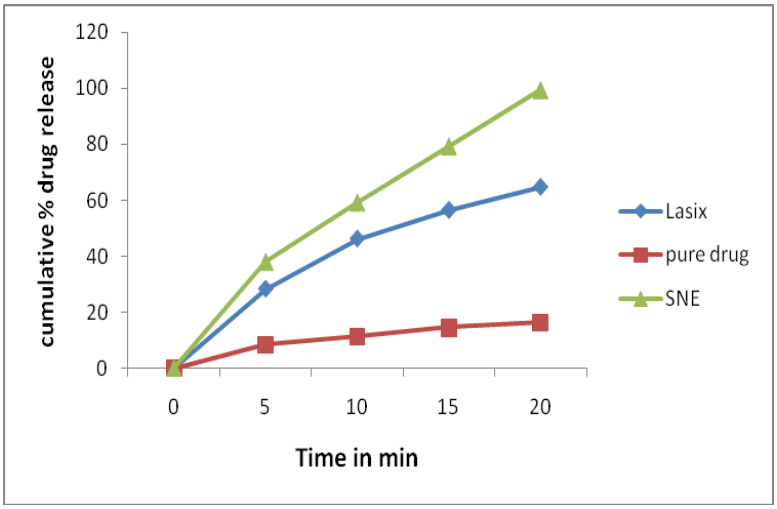

Fig. 5. In–vitro dissolution profile of s-SNEDDS, API and Marketed tablet

3.5.8. Accelerated Stability Testing After 6 months

Accelerated stability study formulation was evaluated for parameters such as effect of Dilution, Droplet size, PDI and In–Vitro drug release. s-SNEDDS passed the test of Effect of Dilution. Droplet size was found to be 17.67 nm with PDI 0.502 indicating no effect on Droplet size after 6 months stability study. Cumulative percentage of Furosemide from s-SNEDDS was 99.51 ± 2.56% at the end of 6 months indicating no change in % drug release after 6 months stability study. Results are given in **Table 9 and Fig. 6**.

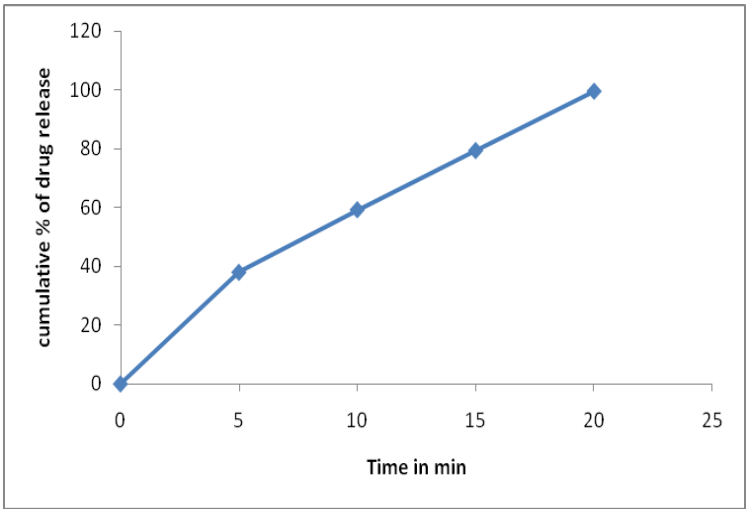

Fig. 6. In–vitro dissolution profile of s-SNEDDS at the end of 6 months accelerated stability study

Table 9: Accelerated stability study of s-SNEDDS after 6 months

Formulation	Effect of dilution	Droplet size (d. nm)	PDI	Drug release (AM ± SD for n=3)
SNE2	Passed	17.67	0.502	99.51 ± 2.56

4. CONCLUSION

A Solid nanoemulsion of a poorly water-soluble drug, furosemide was formulated. The formula composition of liquid SNEDDS was identified based on solubility studies, pseudo ternary phase diagram and droplet size analysis. SNEDDS formulation was converted into s-SNEDDS using aerosil as carrier in 1:2 ratio by adsorption technique. Prepared s-SNEDDS showed “Good” flow properties. Prepared s-SNEDDS showed spontaneous emulsification i.e. in less than 1min with Droplet size 17.42nm and PDI 0.468. From In-Vitro drug release profile it was clear that the percentage drug release from sSNEDDS is 6.1 times and 1.5 times higher than that of pure drug and marketed tablet. Accelerated stability testing at 40°C and 75%RH for solid nanoemulsion of furosemide showed formulation was stable for 6 months.

Hence the present study concluded that s-SNEDDS formulation could be used to improve the solubility and dissolution of furosemide.

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