International Journal of Pharmacy and Biological Sciences
ISSN: 2321-3272 (Print), ISSN: 2230-7605 (Online)
IJPBS | Volume 8 | Issue 1 | JAN-MAR| 2018 | 111-127
Research Article | Biological Sciences | Open Access | MCI Approved| ज्ञान-विज्ञान विमुक्तये
|UGC Approved Journal|

SOLUBILITY ENHANCEMENT OF POORLY SOLUBLE DRUG KETOCONAZOLE BY SELF-EMULSIFYING DRUG DELIVERY SYSTEM

M. Sunitha Reddy*, Yama Narendra[1] and S.Md. Fazal-Ul-Haq[1]

***[1]Centre for Pharmaceutical Sciences, Institute of Science and Technology, Jawaharlal Nehru Technological University Hyderabad, Kukatpally, Telangana, India.**

***Corresponding Author Email: baddam_sunitha@rediffmail.com**

ABSTRACT

Ketoconazole (Kcz) is an imidazole antifungal drug belongs to the class II of Biopharmaceutical Classification System (BCS). The aim of the present work was to prepare Self emulsifying drug delivery system (SEDDS) of lipophilic anti-fungal! Drug, Ketoconazole for improving its solubility and bioavailability. Different oils, surfactants and co-surfactants were screened for their propriety in the formulation of SEDDS. Based on the solubility studies, Capryol 90was selected as oil phase, Polyethylene glycol400 (PEG) as surfactant, Cremophor RH as Co surfactant. The prepared formulations were evaluated for parameters like drug content, percentage transmittance and centrifugation test. Pseudo ternary stage outlines were developed to decide the Nano emulsion range for every formulation. In order to increase the patient compliance, the liquid SEDDS were converted into solid dosage form by adsorption technique. The optimized formulation was added on to an adsorbent, Neusilin. The drug release from these optimized formulations, Solid Self emulsifying drug delivery systems (SSEDDS) was also studied and found to be better compared to the conventional dosage form. Our studies indicate that S-SEDDS can be adequately formulated by adsorption technique. Ketoconazole SEDDS with enhanced dissolution rate and bioavailability were effectively formulated and evaluated.

KEY WORDS

Capryol, PEG 400, Ketoconazole, Neusilin. Self-Emulsifying Drug Delivery System

INTRODUCTION:

There has been a consistent improves in number of new chemical entities, which possess poor aqueous solubility as a result of current drug discovery techniques, and oral delivery of such drugs is regularly associated with short bioavailability. There is a no. of techniques to beat the problems of very low solubility and bioavailability, which may result in better therapeutic efficacy of these drugs. The techniques like multipart formation with cyclodextrins, solid dispersion, and liposome formation, and micronization, use of micelles, co grind and emulsification have been used for improving the dissolution outline of drugs with low solubility.

Self-emulsifying drug delivery systems (SEDDS) have shown huge importance to enhance the oral bioavailability of lipophilic drugs. SEDDS are isotropic blends of oils, surfactant and co-surfactant. They self-emulsify quickly in the aqueous contents of stomach under the mild digestive motility in the G.I tract to present the drug in solution in little droplets of oil.

Ketoconazole is an antifungal drug, belonging to class II drug in BCS classification. One of the most important issues with this drug is its very low solubility in biological fluids, which outcome in poor bioavailability

after oral administration. The solubility of Ketoconazole in aqueous medium is very low, which leads to poor dissolution and hence difference in bioavailability. Hence the main purpose of the study was to develop and evaluate an optimal SSEDDS of Ketoconazole, to improve its bioavailability and also to attain sustained activity.

MATERIALS AND METHODS

Ketoconazole was a donation sample from Natco pharmaceutical pvt. ltd (Hyderabad) Cremophor Rh (Polyoxyl35 -Castor oil) and Trascutolcapryol, Labrafil were obtained from Gattefosse, Mumbai, Tween80 (Polyoxyethylene Sorbitan Monolaurate), PEG (Polyethylene Glycol400).

METHODS

Solubility Studies:

Solubility studies were carried by placing an excess amount of Ketoconazole in a screw capped vials containing 1gr of vehicles (oils, surfactants and co-surfactants). The suspensions of vehicles were heated on a water bath at 40 °C to facilitate the solubilization using vortex mixer. The suspensions were then continuously agitated on a rotary shaker for 48h at ambient temperature. After reaching equilibrium the samples were centrifuged at 5000 rpm for 15min and the supernatant was taken, filtered through 0.45µm membrane filters. The filtrates were suitably diluted with methanol and analyzed spectrophotometrically for the dissolved drug at 257nm.Blank was prepared by dissolving respective vehicles in methanol with same dilution as for the samples. Concentration of ketoconazole in each of the vehicle was calculated from calibration curve.

Construction of pseudo-Ternary Phase Diagrams:

Pseudo-ternary phase diagrams were constructed to determine the appropriate ratios for selected oil, surfactant, and co-surfactant with water at room Temperature by water titration method. The surfactant poly ethylene glycol (PEG400) was mixed with co-surfactant (Cremophor rh) in ratio 4:1, 1:4, 3:1, 2:1 and 1:1 respectively. Aliquots of surfactant/co-surfactant mixture(Smix) were then mixed with oil (Capryol 90) at ratios of 9:1, 8:2, 7:3, 6:4, 5:5, 4:6, 3:7, 2:8, 1:9 in different vials and then titrated with water and note down the weight of water on each addition at room Temperature. The samples were then equilibrated for 30seconds and visually observed after each addition. Based on visual observation the systems were classified as nano emulsion, micro emulsion and coarse dispersion and gel phases. 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 Nano and Micro emulsions.

Preparation of Liquid SNEDDS:

A series of SNEDDS formulations were prepared with varying ratios of oil (20-40 %), surfactant (30-70%) and co-surfactant (10-50 %). A single dose of KCZ (100 mg) was incorporated in all formulations. The total weight of the formulations was kept at 500 mg. The formulations were prepared by dissolving the drug in oil followed by addition of surfactant and co surfactant in glass vials. The resulting mixtures were stirred continuously by vortex mixer followed by sonication for few minutes to obtain a homogenous isotropic mixture. The SNEDDS formulations were stored at ambient temperatures until further use.

Preparation of Solid Self Nano Emulsifying Drug Delivery System

Adsorption to solid carriers: Free flowing powders may be obtained from liquid SNEDDS formulations by adsorption into solid carriers. The adsorption process is simple and just involves addition of the liquid formulation onto carriers by mixing in a blender. The resulting free flowing powder filled directly into capsules. A significant benefit of the adsorption technique is good content uniformity. SEDDS can be adsorbed onto suitable carriers' Solid carriers such as Neusilin. Solid-SNEDDS was prepared by mixing liquid SNEDDS containing KCZwith Neusilin in 1:2 proportions. In brief liquid SNEDDS was added gradually over the carriers contained in a mortar. After each addition, mixture was mixed vigorously and homogenized to ensure uniform distribution of formulation. Resultant damp mass was passed through sieve no. 120 and dried at ambient temperature and stored until further use.

Characterization of SNEDDS:

Self-emulsification and visual assessment: The prepared emulsions were added drop wise to 250ml of water. Self-emulsifying mixtures should quickly

disperse in water on with mild shaking and observe the formed emulsion by visually.

Dispersibility Test: The Time taken for the formation of Nano emulsion was determined by drop wise addition of 1gr of formulation into 250mL of distilled water, simulated gastric fluid and phosphate buffer of pH 6.8 in separate glass beakers at 37 ^{o}C. The contents were stirred using magnetic stirrer at 100rpm. The tendency to form an emulsion was assessed as "good" when the emulsification occurs rapidly less than 1 minute with clear (or) transparent appearance. The tendency to form an emulsion was assessed as "bad" when there is less clear emulsion formation. Based on visual appearance and time taken for self-emulsification, formulations are graded as,

Grade I: Rapidly forming (within 1min) Nano emulsion having a Clear (or) Bluish appearance.
Grade II: Rapidly forming, slightly less Clear emulsion, having a Bluish white appearance.
Grade III: Fine Milky emulsion that is formed within 2 minutes.
Grade IV: Dull, greyish white emulsion with a slight oily appearance that is slow to emulsify (More than 2 minutes).

Phase separation and stability study of emulsions: Each Selected formulation (50µl) was added to a vial containing 5mL of double distilled water, simulated gastric fluid at room Temperature and cyclo mixed for 1 minute and then each mixture was stored and observed for phase separation and precipitation of drug at intervals 2, 4, 6, 8, 12, 24 hours period of time.

Effect of Dilution:

Selected formulations were subjected to dilution in different ratios of 1:10, 1:50, 1:100 and 1:1000-fold dilution with distilled water, Simulated gastric fluid (pH 1.2) and phosphate buffer (pH 6.8). The diluted emulsions were stored for 24 h and observed for any physical changes such as precipitation or phase separation.

Percentage Transmittance:

Each Selected formulation (100µL) was added to a vial containing 10mL of double distilled water, 0.1 N HCl and phosphate buffer of pH 6.8 at room Temperature and cyclomixed for 1minute. Each sample was observed for %Transmittance at 294nm.

Drug loading efficiency: The amount of drug present in the formulation was determined UV-Spectrophotometrically. 50mg of each formulation was accurately weighed and dilute with 100ml of methanol. Resultant solutions were analyzed spectrophotometrically following suitable dilution. Drug loading efficiency was calculated by following equation:

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

FT-IR Studies:

FT-IR Spectrum of pure drug and drug-excipients were obtained by FT-IR Spectrophotometer (Bruker-Alpha). The spectrums of drug, excipients and Formulation were taken with the accumulation 24 scans and a resolution of 4cm-1 over the range of 400- 4000 cm^{-1}. The spectrum of drug-excipient mixtures so obtained was compared with spectrum of pure drug for any interactions or Incompatibilities.

Thermodynamic stability studies:

The physical stability of a SNEDDS formulation is very important for its performance as its can be adversely affected by precipitation of drug in excipient matrix. Poor physical stability of formulation can lead to phase separation of excipients which affects bioavailability as well as therapeutic efficiency. Also, the incompatibilities between formulation &shell of capsule may cause brittleness, softness, and delayed disintegration or incomplete release of drug. The following cycles was carried out for these studies

Centrifugation: In order to estimate metastable systems, the optimized SNEDDS formulations were diluted with 100 times with distilled water. Which pass heating –cooling cycle are centrifuged at 3500 rpm for 30 min. Those formulations that does not show any phase separation are taken for the freeze thaw stress test.

Freeze thaw cycle:

This test was performed for accelerated stability testing of Nano emulsion formulations. In this study three freez thaw cycle of formulations were exposed between temperatures -20 ^{o}C and +25 ^{o}C for each temperature cycles not more than 48 hrs. after 48hrs samples were observed for phase separation (or)

precipitation. The formulations which showed the maximum stability were selected for further study.

In-vitro drug release study:

The in-vitro dissolution study of SNEDDS which were filled into suitable size capsules and carried out using USP-Type II dissolution test apparatus (DS1800 Lab India) in 900mL of 0.1N HCL (pH 1.2) at 37±0.5 ^{o}C with 100rpm rotating speed. In – Vitro drug release study was performed for 90 mins in 0.1NHCl. 5ml of Samples were withdrawn for each time intervals at 0, 5, 10, 15, 20, 30, 45, 60, 75, 90 minutes time intervals and filtered through 0.45µ filter. An equal volume of fresh dissolution medium was replenished after every sampling to maintain constant volume. Samples were analyzed using UV-Spectrophotometer at 225nm. Percentage drug release and cumulative percentage drug release were calculated from absorbance and concentration that were obtained with the help of standard graph of Ketoconazole.

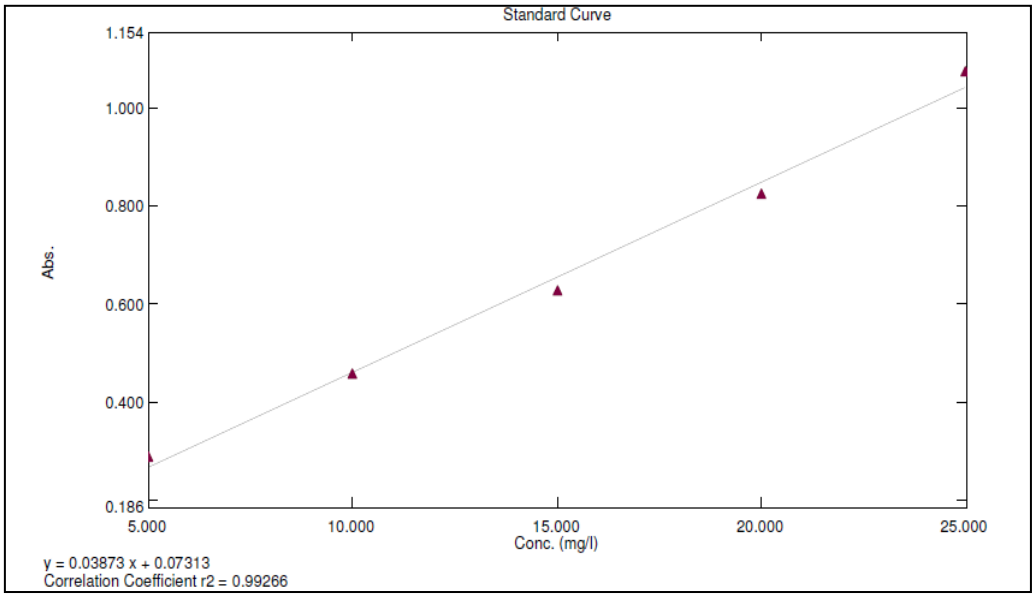
Droplet size and Zeta potential determination: SNEDDS formulations were diluted to 100 times with distilled water in beaker with constant stirring on a magnetic stirrer. The droplet size distributions and Zeta potential of resultant Nano emulsion were determined after 1 hr by Dynamic Light Scattering (DLS) spectroscopy using a Zetasizer Nano ZS Version 6.20 (Malvern Instruments, UK). Size analysis was performed at 25ºc by placing in an electrophoresis cell with an angle of detection of 90ºC for measurement.

RESULTS:

Determination of λ max and Calibration Curve of Ketoconazole in 0.1N HCL:

The spectrum shows a maximum absorption at 220 nm.

Concentration (µg/mL)	Absorbance
5	0.288
10	0.458
15	0.627
20	0.824
25	1.013



Solubility:

Solubility of Ketoconazole (Kcz) in various Oils was determined by UV spectrophotometer the saturation solubility of Ketoconazole in various oils is shown in table Capryol oil was elected for the formulation which forms good.

Table 1: solubility Kcz of various oils

Oil	Solubility
Capryol 90	51.2
Pecol	27.2
Lauroglycol	21.69
Labrafac PG	35.2
Labrafac lipofile	30.3
Soyabean oil	15.2

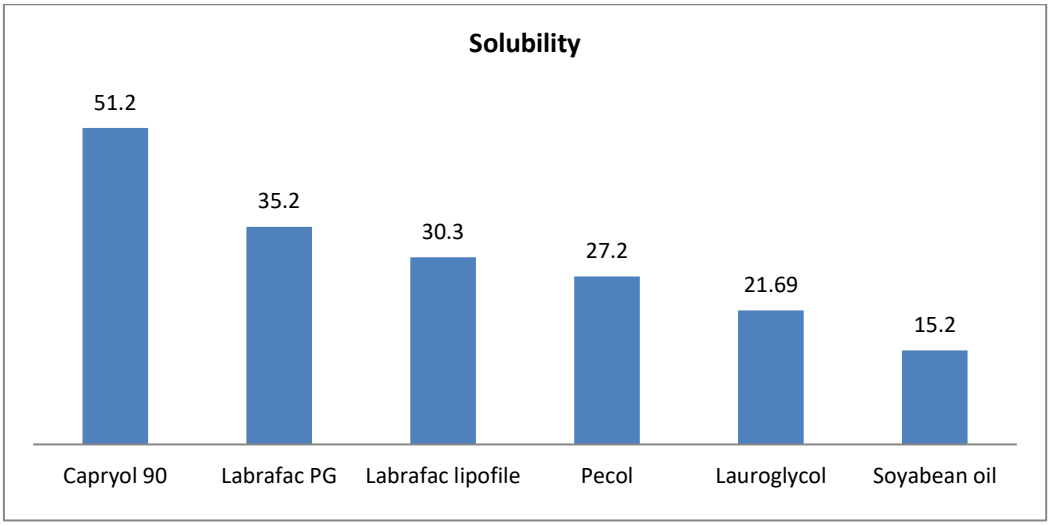


Figure 1: Solubility of Ketoconazole in Various oils

Solubility of Ketoconazole in various Surfactants

Solubility of Ketoconazole was determined in various Surfactants. Surfactants Tween80 is elected for formulation which has highest solubility and good emulsifying ability among all Other formulations.

Table2: Solubility of KCZ in various co-surfactant

Surfactant	Solubility
Tween 80	43.4
span	30.5
labrasol	34.2
PEG400	44.99

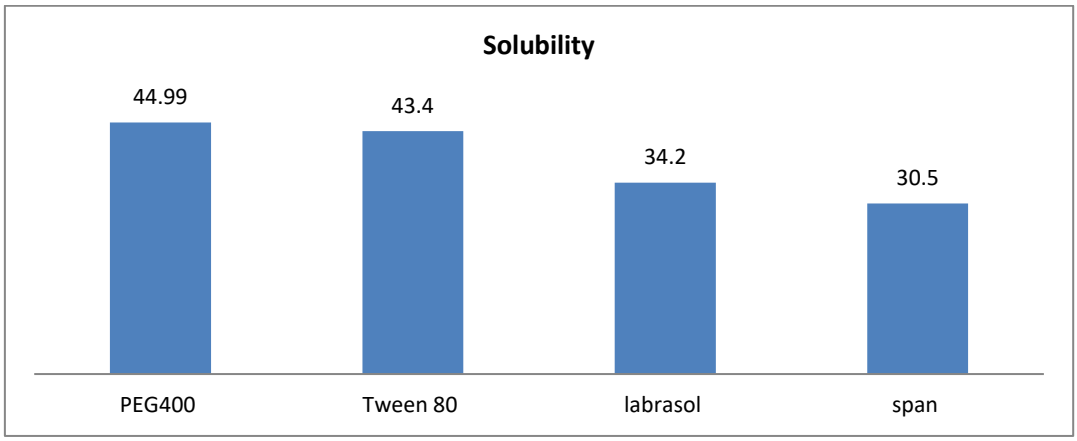


Figure2: Solubility of Ketoconazole in Various Co-Surfactants

Solubility of Ketoconazole in various Co-Surfactants:

Solubility of Ketoconazole in various Co-Surfactants was determined. PEG 400 is select for the formulation which shows highest solubility than other co-surfactants. **Table3:** Solubility's of various Co-Surfactants are shown in Table:

Co surfactant	Concentration
p.glycol	31.5
Cremophor rh	42.6
Trascutol	29.5

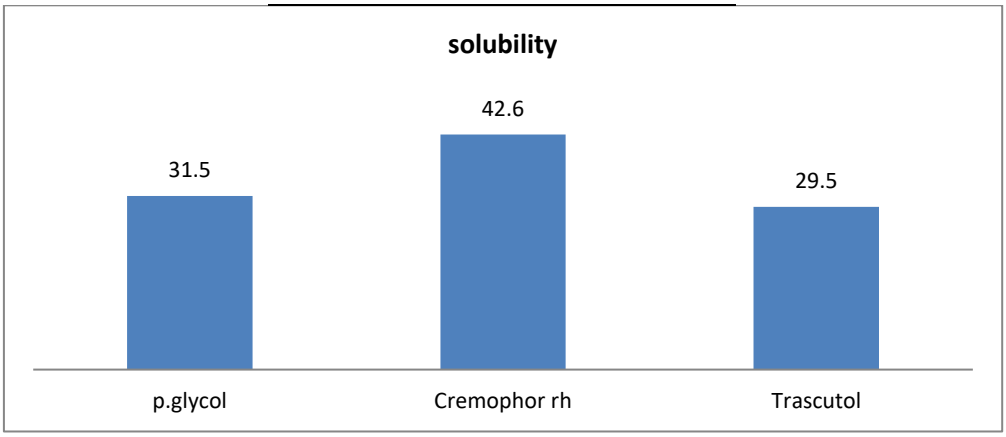


Figure3: Solubility of Ketoconazole in Various Co-Surfactants

Selections of excipients:

Based on the solubility studies done on various oils, surfactants and co surfactants, excipients which have shown more solubility was selected for the formulation.

Oil: Capryol oil

Surfactant: PEG 400

Co surfactant: Cremophore Rh

FT-IR Studies:

The spectrums of drug-excipient mixtures and the formulations so obtained were compared with spectrum of pure drug for any interactions. Characteristic peaks were observed at 3406.5 $cm^{-1,}$ 2874.4cm^{-1}, 2350.1 cm^{-1}, 1640.3 cm^{-1}, 12093 cm^{-1} , 634.4 cm^{-1} for NH stretching vibration, OH stretching, C=N stretching, c=c stretching, and bending of C-CL groups respectively.

FT-IR spectrum of pure drug and the formulation were almost related because of same functional groups. It indicates there was no interaction between Ketoconazole and excipients used in formulation.

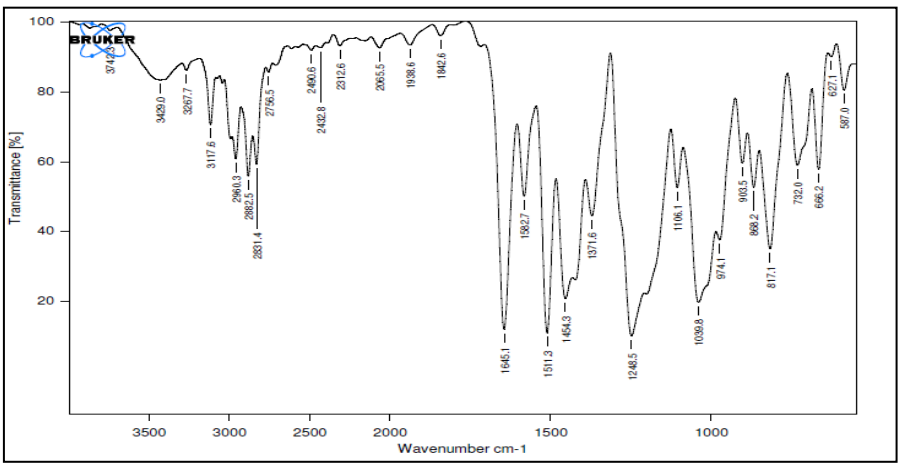


Figure4: FT-IR Spectra of pure drug (Ketoconazole)

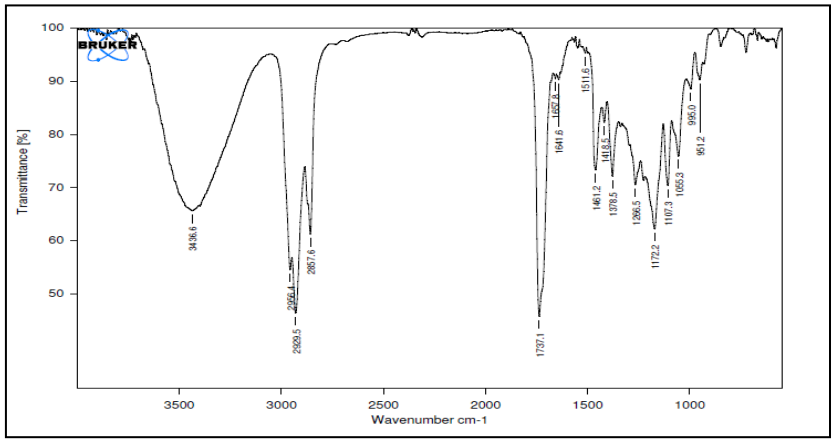


Figure5: FT-IR Spectra of Capryol oil

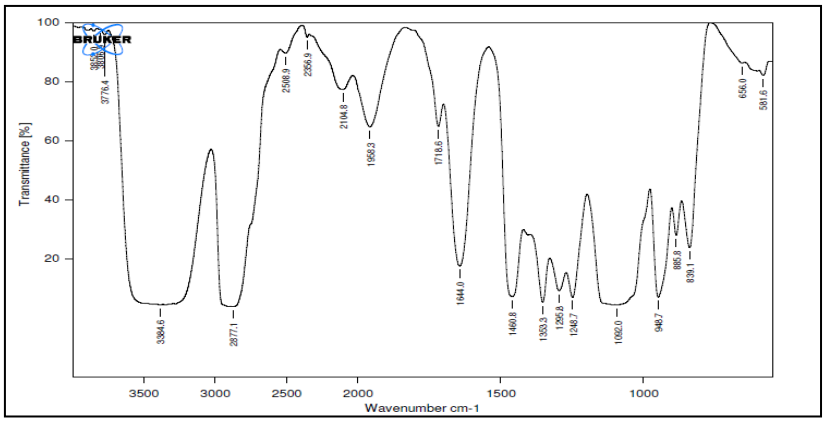


Figure 6: FT-IR Spectra of PEG400

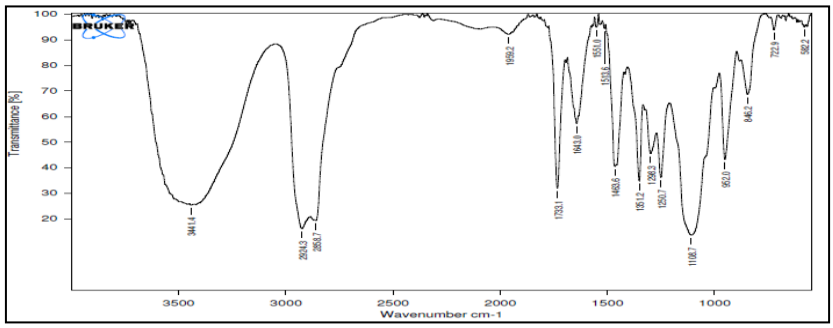


Figure7: FT-IR Spectra of Cremophor Rh

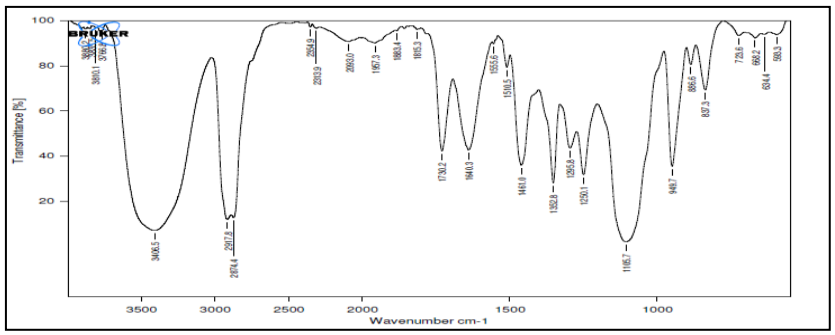


Figure8: FT-IR Spectrum of liquid SNEDDS

Pseudo – ternary Phase Diagrams are constructed to classify the nano & micro emulsion regions and to classify suitable composition of oils, surfactants and co-surfactants for formulation of SNEDDS. From Pseudo – ternary phase diagrams it has been found that the systems consisting of Capryol oil as oily phase,

PEG400 as surfactant and Cremophor RH as co-surfactant showed good nano emulsifying property though drug has been shown more solubility in systems containing Capryol oil as oil phase, tween as surfactant and PEG 400 as co surfactant based on solubility studies. It was also found that systems containing tween as surfactant showed appearance of micro emulsion.

For S_{mix} 4:1 ratio formulation CAPEGCR41showed bluish transparent Emulsion (BTE) for Oil: S_{mix} 1:9,2:8; formulation CAPGCH41 showed Milky white Emulsion (MWE) for Oil:S_{mix} (3:7, 4:6, 5:5, 6:4, 7:3, 8:2, 9:1).

For S_{mix} 3:1 ratio formulation CAPEGCR31 showed bluish transparent emulsion (BTE) for Oil:S_{mix} (1:9, 2:8, 3:7, 4:6, 5:5, 6:4, 9:1and milky white emulsion for 3:7, 4:6, 5:5, 6:4, 7:3, 8:2 ,9:1)

For S_{mix} 1:4 ratio formulations CAPEGCR14 showed clear transparent Emulsion (CTE) for Oil:S_{mix} (1:9, 2:8, 3:7, 4:6) and milky white emulsion for (5:5, 6:4, 7:3, 8:2 ,9:1)

For S_{mix} 4:1 ratio formulations CATCR41 clear transparent Emulsion (CTE) for Oil:S_{mix} 1:9 and milky white emulsion for (5:5, 6:4, 7:3, 8:2 ,9:1)

It was also found that for systems consisting of Capryol oil, PEG, Cremophore Rh, by increasing co-surfactant proportion in S_{mix} systems had shown decreasing property of forming nano emulsion. From this observation it is also clear that Surfactant is playing role to form nano emulsion in a proper range.

Nano emulsion region that is observed in the formulations has been shown Figures (a, b, c & d). Percentage composition of Oil, S_{mix} and water consumed during titration.

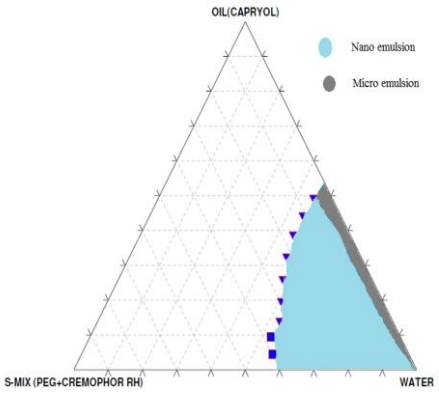


Figure A

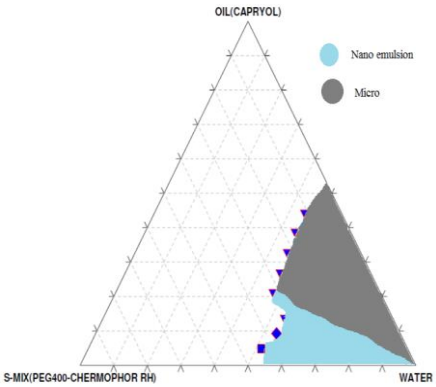


Figure B

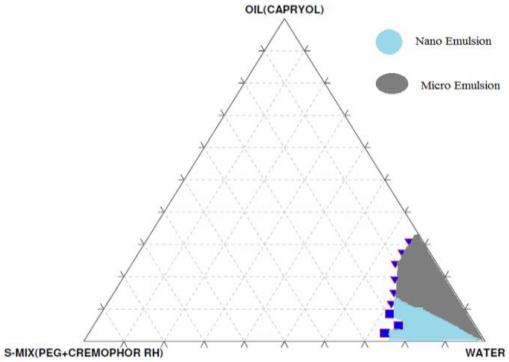


Figure C

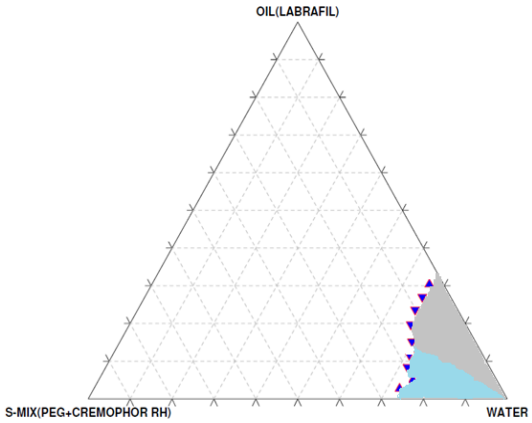


Figure D

Pseudo – ternary Phase Diagrams

A) CAPEGCR4:1, B) CAPEGCR3:1, C) CAPEGCR1:4, D) LFPEGCR11:4

SIZE AND POTENTIALDETERMINATION:

Prepared formulations are analyzed in zeta sizer for the determination of size and potential values. They are shown in various tables below

Table4: Formulation: CAPEGCR31

Oil:S_{mix}	Size of emulsion Droplets (dnm)	Region	Zeta potential	PDI
CAPEGCR41 1:9	72.55	Nano	-10.2	0.643
CAPEGCR41 2:8	38.41	Nano	-6.80	0.120
CAPEGCR 41 3:7	56.41	Nano	-1.476	0.476
CAPEGCR 41 4:6	38.08	Nano	-1.048	0.441
CAPEGCR 41 5:5	44.40	Nano	-0.7	0.834
CAPEGCR 41 6:4	85.7	Nano	-0.4	0.765
CAPEGCR 41 7:3	92.3	Nano	-2.2	0.923
CAPEGCR 41 8:2	99.97	Nano	-1.6	0.687
CAPEGCR 41 9:1	106.6	Micro	-1.3	0.832

Table5: Formulation: CAPEGCR41

Oil:S_{mix}	Size of emulsion Droplets (dnm)	Region	Zeta potential	PDI
CAPEGCR 311:9	118.6	Micro	-3.60	0.648
CAPEGCR 312:8	32.43	Nano	-10.3	0.138
CAPEGCR 313:7	41.13	Nano	-11.8	0.982
CAPEGCR 314:6	58.41	Nano	-14.4	0.786
CAPEGCR 315:5	112.63	Micro	-18.8	0.845
CAPEGCR 31 6:4	109.6	Micro	-16.4	0.654
CAPEGCR 317:3	122.53	Micro	-12.6	0.623
CAPEGCR 318:2	152.30	Micro	-13.5	0.785
CAPEGCR 319:1	162.03	Micro	-15.7	0.823

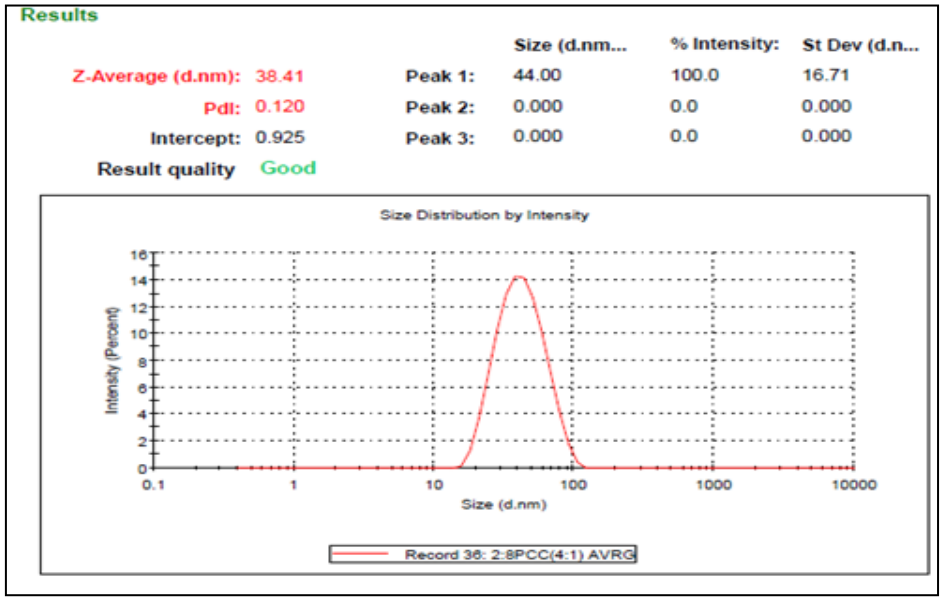


Fig.9: size distribution of CAPEGCR412:8

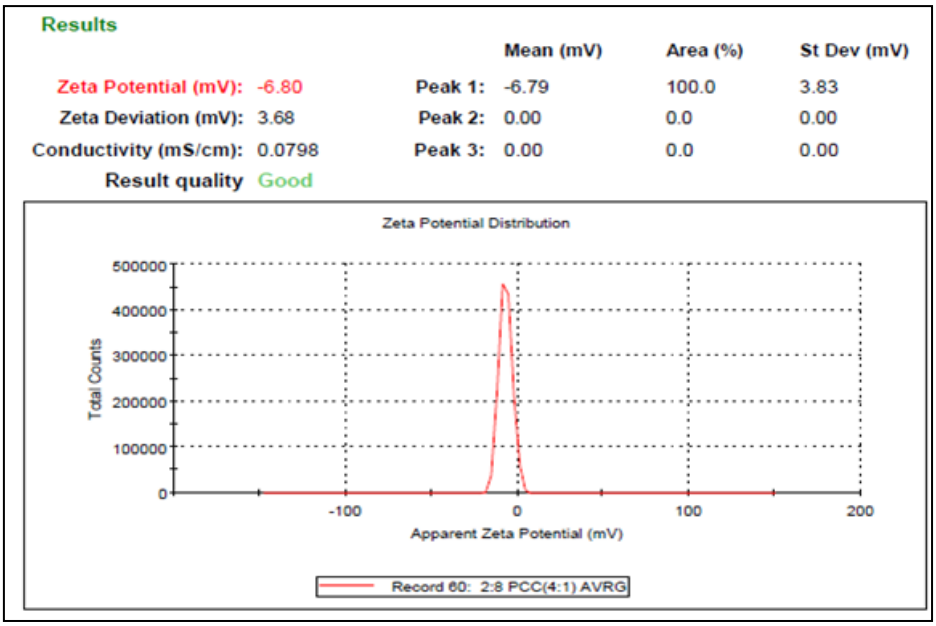


Fig.10: zeta potential of CAPEGCR412:8

EVALUATION TESTS:

Self-emulsification and visual assessment

According to visual assessment formulations are graded for self-emulsification time. Self-emulsifying mixtures should disperse readily in aqueous medium with mild shaking. Self-emulsification time that was determined for prepared SNEDDS are given in Table. The prepared SNEDDS of Ketoconazole were emulsified less than 1min (20-32 sec). Efficiency of all prepared emulsions was good.

Table6: Self emulsification and visual assessment

S.No	Formulation	Emulsification Time	Remark
1	CAPEGCR41 2:8	24±1.2 sec	Good
2	CAPEGCR41 3:7	28±1.5 sec	Good
3	CAPEGCR31 2:8	26±1.5 sec	Good
4	CAPEGCR14 3:7	28±1.8 sec	Good

DISPERSIBILITY TEST:

The five formulations showed grade 1 emulsions when the test is performed in distilled water, 0.1NHCl and phosphate buffer 6.8.

Table7: Dispersibility Test Results

S.No	Formulation name	Distilled water	0.1NHCL	Phosphate buffer of pH6.8
1	CAPEGCR 412:8	Grade 1	Grade 1	Grade1
2	CAPEGCR 413:7	Grade 1	Grade 1	Grade1
3	CAPEGCR 312:8	Grade 1	Grade 1	Grade 1
4	CAPEGCR 143:7	Grade 1	Grade 1	Grade 1

Phase separation and stability study of emulsions

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

Table8: Phase separation and precipitation of the drug

S.No	Formulation	Precipitation	Phase separation
1	CAPEGCR 412:8	No	No
2	CAPEGCR 413:7	No	No
3	CAPEGCR 312:8	No	No
4	CAPEGCR 143:7	No	No

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.

Table 9: Robustness to Dilution Results

S.No	Formulation Name	Distilled Water	0.1N HCL	Phosphate Buffer of Ph6.8
1	CAPEGCR 412:8	No	No	No
2	CAPEGCR 413:7	No	No	No
3	CAPEGCR 312:8	No	No	No
4	CAPEGCR 143:7	No	No	No

Percentage Transmittance

Each diluted sample was observed for % Transmittance at 294nm. Results are given in Table NO 10

All formulations showed %transmittance more than 95% indicating clear emulsions.

Table10: Percentage Transmittance Results

S.No	Formulation Name	Distilled Water	0.1 N HCl	Phosphate Buffer Ph6.8
1	CAPEGCR 412:8	96.3±0.25	98.56±0.78	97.67±0.324
2	CAPEGCR 413:7	97.56±0.98	97.68±0.43	96.3±0.567
3	CAPEGCR 312:8	95.56±0.98	97.56±0.98	98.56±0.98
4	CAPEGCR 143:7	96.56±0.98	95.56±0.98	94.56±0.98

Drug loading efficiency

50mg of each SNEDS formulation was diluted with 100mL Methanol. Resultant solutions are analyzed UV-Spectrophotometrically following suitable dilution. Absorbance of each solution is measured at 294nm. Results are given in Table No 11. It was found both formulations have drug loading efficiency more than 90%.

Table11: Drug loading efficiency of formulations

S.No	Formulation Name	Drug Loading Efficiency
1	CAPEGCR 412:8	98.34±0.678
2	CAPEGCR 413:7	96.59±0.56
3	CAPEGCR 312:8	97.46±0.66
4	CAPEGCR 143:7	95.56±0.16

Thermodynamic stability studies

Thermodynamic stability study is designed to identify Metastable formulation. The SNEDDS are subjected to Centrifugation study & 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

Table12: Thermodynamic stability studies

S.No	Formulation name	Centrifugation (3,500rpm for 30min)	Freeze thaw cycle (-20 °C and +25 °C)
1	CAPEGCR 412:8	*P	*P
2	CAPEGCR 413:7	*P	*P
3	CAPEGCR 312:8	*P	*P
4	CAPEGCR 143:7	*P	*P

***IN VITRO* DRUG RELEASE STUDY:**

After performing the drug release study for 90 min in 0.1 N HCl pure drug showed the % drug release of 58.21% and the CAPEGCR41 (2:8) showed 94.2%drug release and different formulations showed %drug table.

Table13: cumulative %release of pure drug and formulations:

TIME	Pure drug	CAPEGCR 412:8	CAPEGCR 41(3:7)	CAPEGCR 31(2:8)	CAPEGCR 14(3:7)	LFPEGCR 141:9
0	0	0	0	0	0	0
5	28.21	41.56	39.32	40.32	30.21	39.98
10	36.14	46.25	42.31	42.23	33.21	41.01
15	42.18	51.32	48.55	49.23	38.2	48.32
30	45.98	57.63	51.02	50.23	41.99	51
45	52.69	64.12	55.23	51.23	47.32	53.21
60	54.17	79.36	62.23	58.32	54.23	56.98
75	56.23	84.23	74.21	66.32	62.23	61.99
90	58.21	94.2	81.33	78.98	75.32	74.23

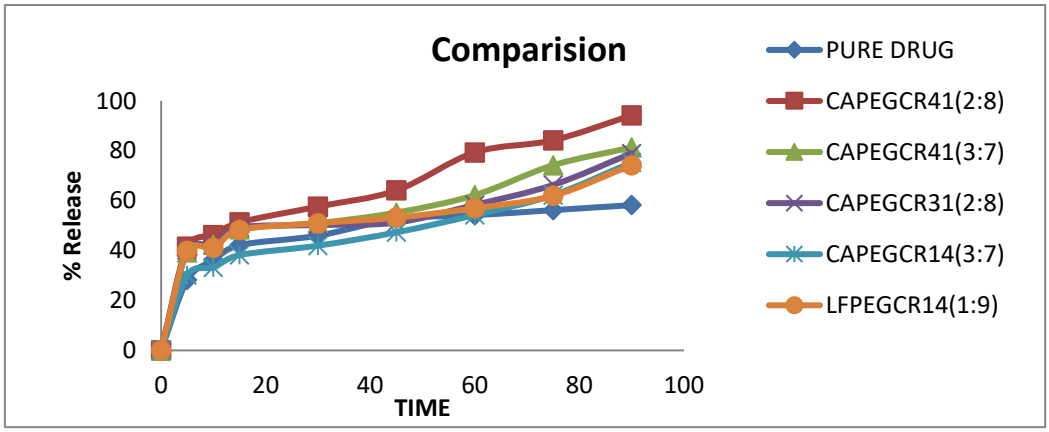


Figure11: cumulative %release of pure drug and formulations.

Droplet size and Zeta potential determination:

From the dissolution study it was concluded that the formulation CAPEGCR41 (2:8) showed more % drug release than other formulation. So, it is selected as the best formulation. Size and potential values are determined for the formulation. Size of the particles was found to be 38.41nm and pdi value was found to be 0.120 and its zeta potential was found to be -6.1mv.

Table14: size, pdi&zeta potential of optimized formulation

S.No	Formulation name	Average droplet size (d.nm)	PDI	Zeta potential (mV)
1	CAPEGCR41 (2:8)	38.41nm	0.12	-6.1mv.

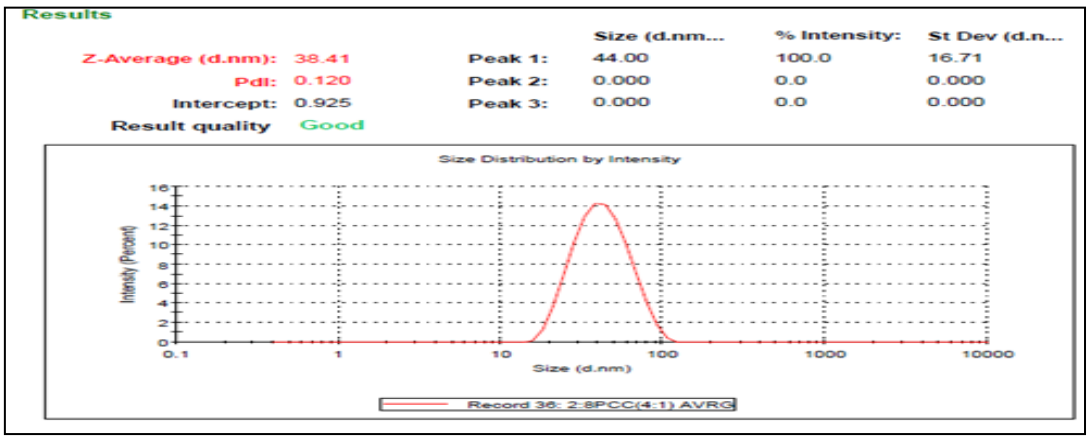


Figure12: size distribution of selected formulation CAPEGCR412:8

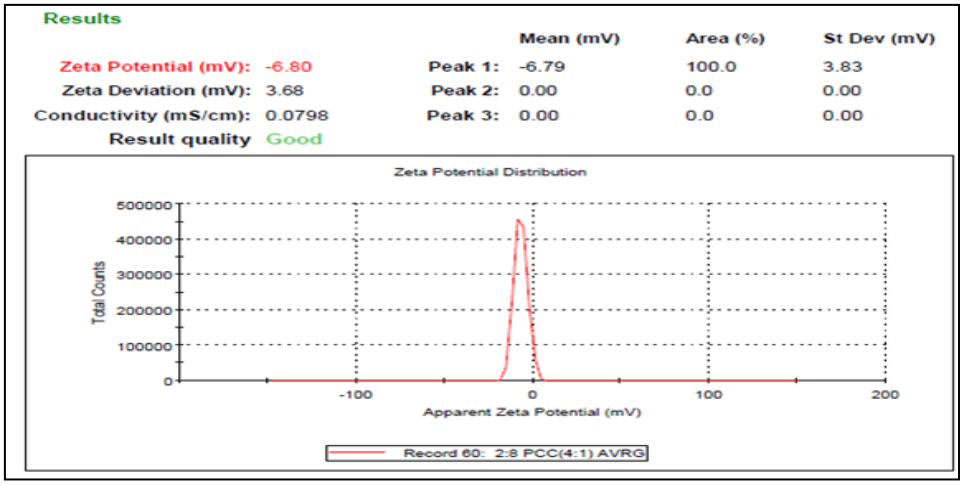


Figure13: zeta potential of CAPEGCR412:8

Preparation of Solid SNEDDS of Ketoconazole:

Based on evaluation tests done for five liquid SNEDDS formulations the CAPEGCR41 (2:8) formulation is selected for preparation of solid SNEDDS of Ketoconazole. Compared to other formulations CAPEGCR41 (2:8) showed good self-emulsification property which was emulsified spontaneously in 24±1.2 sec and also droplet size (36.42 nm) was less than other formulations with more uniform distribution of particles (PDI =0.184). Hence the optimum composition for preparation of s-SNEDDS was found to be capryol oil (28.71%w/w), PEG400 (16.745%w/w), Cremophore (50.235%w/w) and Drug (4.306%w/w). With selected optimum formulation s-SNEDDS are prepared using Neusilin as carrier in 1:2 ratios by adsorption technique.

Evaluation of solid SNEDDS of Ketoconazole:

Table15: Flow properties of s-SNEDDS of Ketoconazole:

Flow Properties	Results
Angle of repose	27.926 ± 1.205
Bulk density (g/mL)	0.375 ± 0.015
Tapped density (g/mL)	0.421 ± 0.015
Compressibility index (%)	10.92 ± 0.05
Hausner's ratio	1.122 ± 0.0067

All values are expressed as Mean ± SD (n=3)

Drug Content:

Amount of drug present in prepared s-SNEDDS was determined. Drug content of the S-SNEDDS was found to be **95.652 ± 1.57 %.**

FT – IR Studies:

The spectrums of drug-excipient mixtures and the solid formulation of Ketoconazole so obtained were compared with spectrum of pure drug for any interactions.

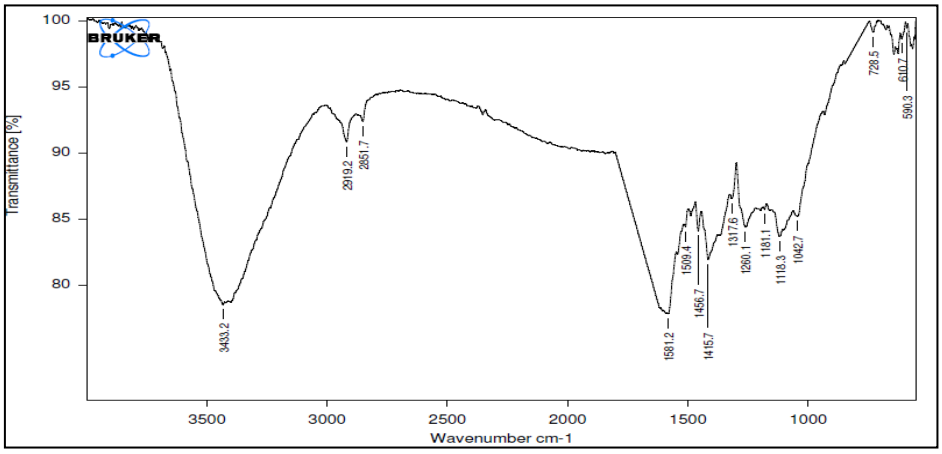


Figure14: FT–IR Spectrum of NEUSILIN

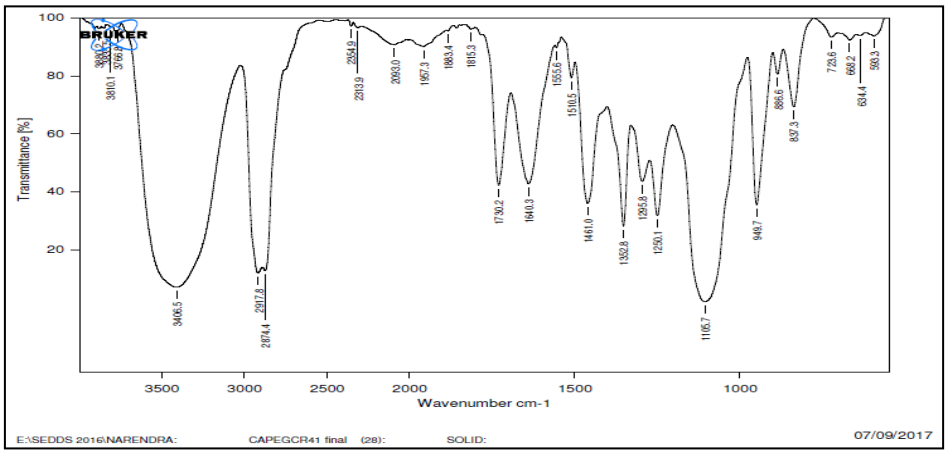


Figure15: FT–IR Spectrum of S-SNEDDS

Table16: Interpretation of IR spectrum of pure Ketoconazole and solid formulation

Functional Group	Ketoconazole (KCZ)	KCZ S-SNEDDS
C=O	1645.1	1640.3
N-H stretching	3267.6	3406.5
OH stretching	2831.4	2874.4
C=N	2312.5	2350.1
C=C	2065.4	2093
C-CL	627.1	634.4

Table17: *In Vitro* dissolution of s-SNEDDS, API and Marketed Product:

Time	Pure Drug	Solid Formulation	Marketed Product
0	0	0	0
5	28.21	39.2	32.23
10	36.14	43.1	38.29
15	43.18	50.23	44.99
30	48.98	53.41	50.53
45	53.69	60.29	58.28
60	56.17	78.11	60.36
75	58.23	82.32	65.36
90	59.21	91.62	70.33

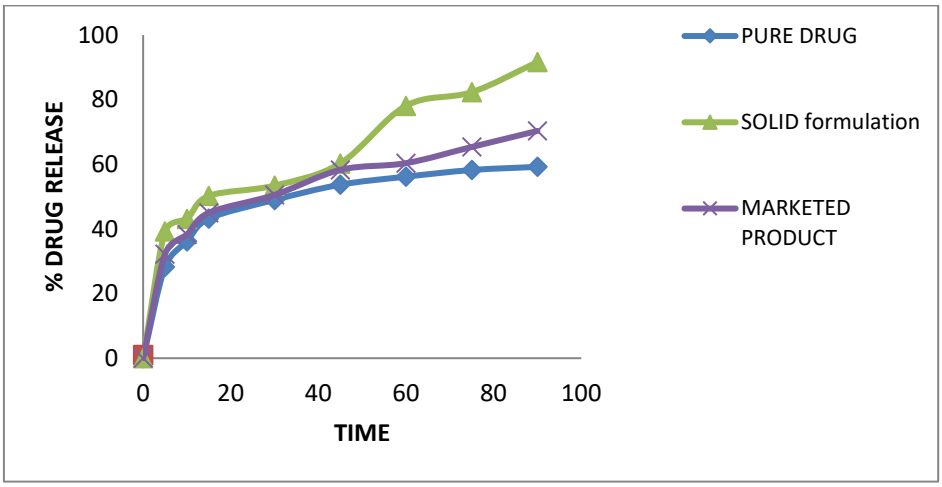


Figure16: *In Vitro* dissolution of s-SNEDDS, API and Mar. Product

DISCUSSION:

In the present work Self Nano Emulsifying Drug Delivery System (SNEDDS) containing a poorly water-soluble drug Ketoconazole were formulated for oral administration. Optimization studies were carried out using various components such as oil, surfactant and co-surfactant. They were further investigated by conducting solubility studies and constructing pseudo ternary phase diagram. Formulation CAPEGCR4:1(2:8) was found optimum in terms of drug loading, rapid emulsification, droplet size and in-vitro release. Solid SNEDDS(S-SNEDDS) of Ketoconazole were also prepared using Neusilin by adsorption process. These formulations showed good flow properties and drug content. Thus, the study confirmed that solid SEDDS formulation can be used as possible alternative to oral formulation of Ketoconazole to improve its solubility and oral bioavailability.

Acknowledgement:

It is proud to express my heartfelt to gratitude to my beloved institutional guide. Centre for pharmaceutical science, IST and JNTUH for their voluble guidance, cooperation, affectionate encourage and moral support throughout the course of this investigation.

REFERENCES:

1. Biopharmaceutical Classification System and Formulation Development. Particle Sciences, 2011; Volume 9.
2. M. Sunitha Reddy*; P.Srinivas Goud; S.S.Apte. Solubility Enhancement of Candesartan Cilexetil By Self Emulsifying Drug Delivery Systems. IJPSR, 2012; Vol.3 (7): 2098-2104.
3. Dr. M. Sunitha Reddy*, Senigarapu Soumya1 and S. Muhammad Fazal Ul Haq1: Solubility Enhancement of Azilsartan By Selfemulsifying Lipid Formulations. Wjpps, 2016; Volume 6, Issue 3, 957-974
4. Mohd Yasir; Mohd Asif; Ashwani Kumar; Abinav Aggarval. Biopharmaceutical Classification System: An Account. International Journal of Pharm Tech Research, 2010; Vol.2, No.3: 1681 – 1690.
5. Kavita Sapra; Ashu Sapra; S K Singh; Saloni Kakkar. Self-Emulsifying System: A Tool in Solubility Enhancement of Poorly Soluble Drugs. Indo Global Journal of Pharmaceutical Sciences, 2012; 2(3): 313 – 332.
6. Eman Atef, Albert A. Belmonte, Formulation and In Vitro and In Vivo Characterization of A Phenytoin Self-Emulsifying Drug Delivery System, European Journal of Pharmaceutical Sciences 35 (2008) 257–263.
7. D.M.Brahmankar; Sunil B. Jaiswal : Biopharmaceutics And Pharmacokinetics A Treatise:345 – 357.
8. WWW.Drugbank.Ca
9. Martin's Physical Pharmacy And Pharmaceutical Sciences – Sixth Edition – Patrick J.Sinko: 48 – 52; 461 – 466.
10. More H.N; Hazare A.A: Practical Pharmaceutics (Physical Pharmacy) Manas Prakashan, Kolhapur, First Edition, 2004; 86 -105.
11. M.G.Wakerly; CW Pouton; B.J.Meakin ; F.S.Morton. Self-Emulsification of Vegetable Oil Non- Ionic Surfactant Mixtures. ACS Symp.Ser.1986; 311: 242 – 255.

12. European Pharmacopeia Fifth Edition: 2413 – 2414.
13. Indian Pharmacopeia.
14. Anusha Reddy Ravula, *Vijaykumar Nagabandi, Shruthi Parney, Encapsulation of Self Emulsifying Drug Delivery Systems (Sedds) Of Lercanidipine Hydrochloride into Hard Gelatin Capsules, International Journal Of Biopharmaceutics. 2014; 5(2): 73-82.
15. www.Gattefosse. Com
16. Raymond C Rowe; Paul J Sheskey; Marian E Quinn. Hand Book of Pharmaceutical Excipients Sixth Edition; 208 – 210, 466 – 467, 748 – 749.

***Corresponding Author:**
M. Sunitha Reddy*
Email: **baddam_sunitha@rediffmail.com**