International Standard Serial Number (ISSN): 2319-8141
International Journal of Universal Pharmacy and Bio Sciences 3(2): March-April 2014
INTERNATIONAL JOURNAL OF UNIVERSAL PHARMACY AND BIO SCIENCES
*IMPACT FACTOR 1.89****
*ICV 5.13****
Pharmaceutical Sciences **RESEARCH ARTICLE……!!!**

FORMULATION & IN-VITRO EVALUATION OF SELF-MICROEMULSIFYING DREUG DELIVERY SYSTEM OF CARVEDILOL

Darji Sweta V.
Department of Pharmaceutics, 252-Shri B.M.Shah College of Pharmaceutical Education and Research, Modasa – 383315, Gujarat.

KEYWORDS:

SMEDDS, Carvedilol, bioavailability, *In-vitro* release study, Zeta potential, Phase diagram.

For Correspondence:
Darji Sweta V.*
Address:
22, Mattruchaya Gayatri Society Meghraj At. Post Ta.-Meghraj,
Pin code- 383350.
Email id: Sweta8darji@gmail.com

ABSTRACT

Purpose: Carvedilol is poorly water soluble drug. It should be come into the BCS Class II drug. Hence oral Bioavailability of Carvedilol is less (25%). To develop novel dosage form of the self-Microemulsifying drug delivery systems (SMEDDS) for the Carvedilol for enhancing its solubility. **Method:** Before the formulation of SMEDDS solubility study was performed in different excipients and select excipients on basis of solubility of Carvedilol. Microemulsion region was decided by preparing ternary phase diagram. Drug excipients interaction study performed FTIR. After preliminary study, SMEDDS formulation was prepared in Capmul MCM NF (oil), Acrysol k-160(surfactant), Transcutol CG (co-surfactant) by simple mixing at 40°C. Parameters evaluated like: macroscopic evaluation, visual assessment, self-emulsification, transmittance test, particle size distribution, zeta potential and polydespersibility index and *In vitro* dissolution. *In vitro* dissolution was carried in USP apparatus II using 0.1N HCl at 37±0.5°C with 50 rpm rotating speed, drug release measured by spectroscopic method. **Result:** From the solubility study, better solubility was seen in Capmul MCM NF (oil), Acrysol k-160(surfactant), Transcutol CG (co-surfactant). No any drug excipients interaction seen. Optimized formulation S_2 of SMEDDS was observed with smaller droplet size 18.3nm, PDI 0.239 and zeta potential -11.7mV. Formulation was clear after dilution with water. SMEDDS formulation showed complete release in 60 minutes as compared with marketed (12.5 mg) tablet. **Conclusion:** SMEDDS Carvedilol oral formulation was prepared that provides excellent drug solubilisation and improved in vitro release of Carvedilol.

INTRODUCTION:

As a consequence of modern drug discovery techniques, there has been increase in the number of new pharmacologically active lipophillic compound that are poorly water soluble. It is great challenge for pharmaceutical scientist to convert those compound into administered formulation with sufficient bioavailabilit.[1] When drug administered through oral route the first step for it to get absorbed is its solubilization followed by its permeation. So, bioavailability problems are produced. These drugs are classified as class II drug by Biopharmaceutical Classification System (BCS), drugs with poor aqueous solubility and high permeability. Oral delivery of such drug is complicated because of their low bioavailability, high intra- and inter-subject variability, and not have dose linearity.[2] To overcome these problems, varieties of strategies have been developed like, Solid dispersion, Use of surfactant, Cyclodextrins, Co-solvents, Salt formation, Nano- and micro- suspension, Lipid based formulation, Micro emulsion, SMEDDS, Liposome, Polymeric nanoparticles, Solid lipid nanoparticles.[3]

SMEDDS are defined as isotropic mixtures of natural or synthetic oil, surfactant or alternatively one or more hydrophilic solvent and co-solvent that have unique ability of forming fine o/w micro emulsion upon mild agitation followed by dilution in aqueous media such as GI fluid. It has a droplet size between 10 – 200 nm. It is transparent in nature than those of conventional emulsion which is opaque. These fine droplets disperse readily in GIT that provide large surface area for the drug absorption with minimum GIT irritation. Due to its small globule size the micro emulsified drug can be absorbed through lymphatic pathways thereby by-passing the hepatic first-pass effect. These are stable preparation. SMEDDS improve the dissolution of the drug due to increased surface area on dispersion and solubility effect of surfactant. The basic difference between self-emulsifying drug delivery systems (SEDDS) also called as self-emulsifying oil formulation (SEOF) and SMEDDS is SEDDS typically produce opaque emulsions with a droplet size between 100 and 300 nm while SMEDDS form transparent micro emulsions with a droplet size of less than 50 nm also the concentration of oil in SMEDDS is less than 20 % as compared to 40-80% in SEDDS. When compared with emulsions, which are sensitive and metastable dispersed forms, SMEDDS are physically stable formulations that are easy to manufacture. Thus, for lipophilic drug compounds that exhibit dissolution rate limited absorption, these systems may offer an improvement in the rate and extent of absorption and result in more reproducible blood-time profiles. SMEDDS formulation is in theory, comparatively simple. The key step is to find a suitable oil surfactant mixture that can dissolve the drug within the required therapeutic concentration. The SMEDDS mixture can be filled in either soft or hard

gelatin capsules. A typical SMEDDS formulation contains oils, surfactants and if required an antioxidants. Often co-surfactants and co-solvents are added to improve the formulation characteristics. [4.5.6.7]

Carvedilol is a non-selective beta blocker. It has been used extensively in patients with hypertension and has also been used in patients with angina and congestive cardiac failure. Oral bioavailability of Carvedilol is very law (25-35%), due to its poor water solubility (log P 4.115). The aim of this investigation was to Formulation & in-vitro Evaluation of SMEDDS formulation of Carvedilol for enhancing its solubility.

2. MATERIAL AND METHOD

2.1 Materials

Carvedilol was obtained as a gift sample from Torrent Pharma Ltd., Ahmedabad, India. The following materials were gifted by Abitec corp., USA, and were used as received: Capmul® MCM NF, Capmul MCM, Captex 200P, Capmul PG 8 NF. Transcutol® P, Transcutol CG, Labrasol were received as gift sample from Gattefosse, France. Acrysol® K 160, Acrysol K® 140, Acrysol® El 135 (Polyoxyl 35 castor oil) were also gifted from Corel Pharma chem., Ahmedabad, India. Tween® 80 (polyoxyethylene sorbitan monooleate), Tween® 60 (Polyoxyethylene sorbitan monostearate), Propylene glycol were bought from Finar Chemical Limited, Ahmedabad, India. Polyethylene glycol 200 (PEG 200), Polyethylene glycol 400 (PEG 400) were bought from S. D. Fine Chemical Limited, Mumbai, India. Sunflower oil, Castor oil was purchased from Gujarat Glycol Private Limited, Ankaleshwar, India and Kush Proteins Private Limited, Anand, India. Double distilled water was used throughout the study. All other chemical were of reagent grade.

2.2 Solubility Studies

3 gm of selected vehicles was added to each cap vial containing an excess of Carvedilol. After sealing, mixing of the systems was performed using a vortex mixer. Formed suspensions were then shaken with a shaker at 25ºc for 48 hours. After reaching equilibrium, each vial was centrifuged at 2000 rpm for 15 minutes. Excess insoluble carvedilol was discarded by filtration using 0.45µm filter disk. Filtered solution was appropriately diluted with methanol and UV absorbance was measured at 224 nm. Concentration of dissolved drug was determined using standard equation.

2.3 Ternary Phase diagram study

On the basis of the solubility study of drug, oil, surfactants, co-surfactants and aqueous phase were used for construction of phase diagram. Oil, surfactant, and co-surfactant are grouped in four different combinations for phase studies. Surfactant and co-surfactant (Smix) in each group were mixed in different weight ratio (1:0, 1:1, 1:2, 2:1, 1:3, 3:1, 1:4, 4:1 etc.). These Smix ratios are chosen in increasing concentration of surfactant with respect to co-surfactant and in increasing concentration of co-surfactant with respect to

surfactant for detail study of the phase diagram for formulation of Nano/micro emulsion. For each phase diagram, oil, and specific Smix ratio are mixed thoroughly in different weight ratio from 1:9 to 9:1 (0.5:9.5, 1:9, 1.5:8.5, 2:8, 2.5:7.5, 3:7, 3.5:6.5, 4:6, 4.5:5.5, 5.0:5.0, 5.5:4.5, 6:4, 6.5:3.5, 7:3, 7.5:2.5, 8:2, 8.5:1.5, 9:1, 9.5:0.5) in different glass vials. Different combination of oils and Smix were made so those maximum ratios were covered for the study to delineate the boundaries of phase precisely formed in the phase diagrams. Pseudo-ternary phase diagram was developed using aqueous titration method. Slow titration with aqueous phase is done to each weight ratio of oil and Smix and visual observation is carried out for transparent and easily flowable o/w Nano/micro emulsion. The physical state of the Nano/micro emulsion was marked on a pseudo-three-component phase diagram with one axis representing aqueous phase, the other representing oil and the third representing a mixture of surfactant and co-surfactant fixed weight ratios.

2.4 Preparation of SMEDDS formulation

The formulations were prepared by initially dissolving required quantity of carvedilol in oil. Then Surfactant and Co-surfactant mixer were added and the final mixture was mixed by vortexing until a clear solution was obtained. The formulation was equilibrated at ambient temperature for at least 48 hr, and examined for signs of turbidity or phase separation prior to self-emulsification and particle size studies. Final formulation was filled in hard gelatin capsule (Size 00).

Table 2. SMEDDS formulation of Carvedilol

Ingredient	Batch (in mg)					
	S_1	S_2	S_3	S_4	S_5	S_6
Capmul MCM NF	200	250	300	200	250	150
Acrysol K-160	400	375	350	480	450	510
Transcutol CG	400	375	350	320	300	340
Carvedilol (mg)*	12.5	12.5	12.5	12.5	12.5	12.5
* Amount of drug in 1 gm SMEDDS Formulation						

Table 4.4 showed six different SMEDDS formulation of Carvedilol. In each formulation amount of drug was 12.5 mg. Total 1 gm formulation were prepared. In the study, different oil to surfactant/co-surfactant ratio were used to check the various parameter of the SMEDDS formulation and from that optimized formula was find out.

2.5. Determination of self emulsification time[8]

The emulsification time of SMEDDS was determined using dissolution apparatus 2. 300 mg of each formulation added drop wise to 500ml purified water at 37°C. Gentle agitation was provided by a standard stainless steel dissolution paddle rotating at 50 rpm. Emulsification time was assessed visually.

2.6. Transmission test[8]

Stability of optimized microemulsion formulation with respect to dilution was checked by measuring transmittance through U.V. Spectrophotometer (U.V.-1800,Shimadzu). Transmittance of samples was measured at 650 nm and for each sample three replicate assays were performed.

2.7. Drug Content[9]

Carvedilol from SMEDDS formulation was extracted in methanol using sonication technique. The solutions were filtered, using wattman filter paper. The methanolic extract was analyzed for the Carvedilol content spectrophotometrically (UV–1800, Shimadzu, Japan) at 285 nm using standard curve.

2.8 Robustness to Dilution[9]

Robustness to dilution was performed with excess of water, standard phosphate buffer pH 6.8 and 0.1N HCl (500-900 ml) and was stored for 12 hours and note down whether precipitation or phase separation was found or not.

2.9. Visual assessment[10]

Carvedilol SMEDDS (approximately 0.2 ml) was diluted with purified water (100 ml) and gently stirred with magnetic stirrer. Temperature should be 37°C. The *in-vitro* performance of the formulation was visually assessed using the following grading system

Grade A: Rapidly forming 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, greyish white emulsion having slightly oily appearance that is slow to emulsify longer than 2 minute

Grade E: Formulation, exhibiting either poor or minimal emulsification with large oil globules present on the surface.

2.10. Droplet size measurement and Polydispersibility Index (PDI)[10]

Formulations (S_1 to S_6) each of 1 ml were diluted with 100 ml of water in a volumetric flask. The volumetric flask was inverted twice to ensure complete dispersion of the formulation. After ensuring complete dispersion of the formulation the droplet size of resultant Microemulsion was determined by photon correlation spectroscopy that analyze the fluctuation in light scattering due to the Brownian motion of the droplets as function of time using a Zetasizer Nano Series (Malvern Instruments, DTS Ver.4.10,

Serial No. MAL 500999). Light scattering was monitored at 25°C at 90° angle. Value of droplet size and polydispersibility index are tabulated in table 5.13 and represented in figure 5.10-5.16

2.11. Zeta potential measurement[11]

Zeta potential for microemulsion was determined using Zetasizer HSA 3000 (Malvern Instrument Ltd., U.K.). Samples were placed in clear disposable zeta cells and results were recorded. Before putting the fresh sample, cuvettes were washed with the methanol and rinsed using the sample to be measured before each experiment.

2.12. *In vitro* Dissolution Studies[11]

The *in vitro* drug release of Carvedilol from the optimized SMEDDS was performed using USP dissolution Apparatus II (TDT-08L, Electrolab, Mumbai, India). Hard gelatin capsules, size "00" filled with preconcentrate (equivalent to 12.5mg Carvedilol) were put into each of 900 ml 0.1 N HCl soln. pH 1.2, at 37 ± 0.5°C with a 50 rpm rotating speed. Samples (5 ml) were withdrawn at regular time intervals S(5, 10, 20, 30, 40, 50 and 60 min) and filtered using a 0.45 m filter. An equal volume of the dissolution medium was added to maintain the volume constant. The drug content of the samples was assayed using UV visible spectrophotometric method at 285 nm wavelength. All measurements were done in triplicate.

3. RESULT AND DISCUSSION

3.1 Solubility studies

Table 3.1 : Solubility of Carvedilol in different oil, surfactant and co-surfactant

Vehicle	Solubility in mg/gm	Function in SMEDDS
Capmul MCM NF	**82.66**	oil
Captex 2P	8.96	Oil
Capmul PG-12	8.13	Oil
Labrafac	5.25	Oil
Capmul PG-8-NF	38.66	Oil
Sunflower oil	3.78	Oil
Castor oil	5.22	Oil
Olive oil	4.32	Oil
Soyabean oil	7.68	Oil
Cottonseed oil	17.12	Oil
Seasam oil	19.33	Oil
Linseed oil	22.61	Oil
Penut oil	8.13	Oil
Labrasol	134.66	Surfactant
Acrysol EL-135	52.66	Surfactant
Acrysol K-150	71.16	Surfactant
Acrysol K-160	**83.46**	Surfactant
Tween 20	50.85	Surfactant
Tween 80	30.12	Surfactant

Span 80	42.28	Surfactant
Transcutol CG	**125.23**	Co-surfactant
PEG 400	100. 28	Co-surfactant
PEG 200	85. 46	Co-surfactant
Propylene glycol	15.04	Co-surfactant
Transcutol P	70.02	Co-surfactant

As per solubility data of Carvedilol in different oils, maximum amount of drug dissolved in Capmul MCM NF. So, It was selected. (82.66 mg/gm)

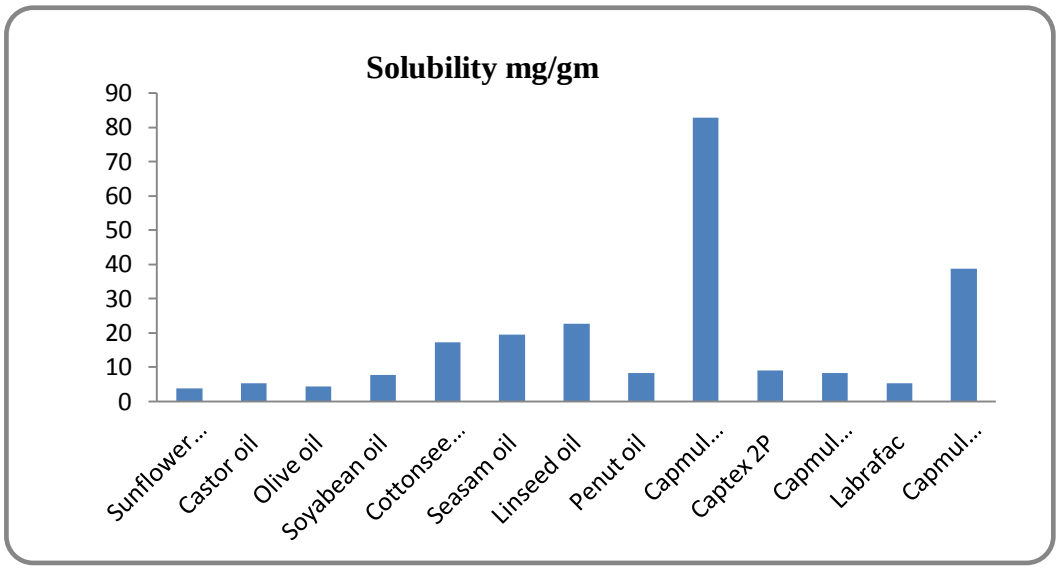


Figure 3.1: Schematic diagram of drug solubility in different oils

As per solubility data of Carvedilol in different surfactant, maximum amount of drug dissolved in Acrysol k-160. So, It was selected. (83.46 mg/gm)

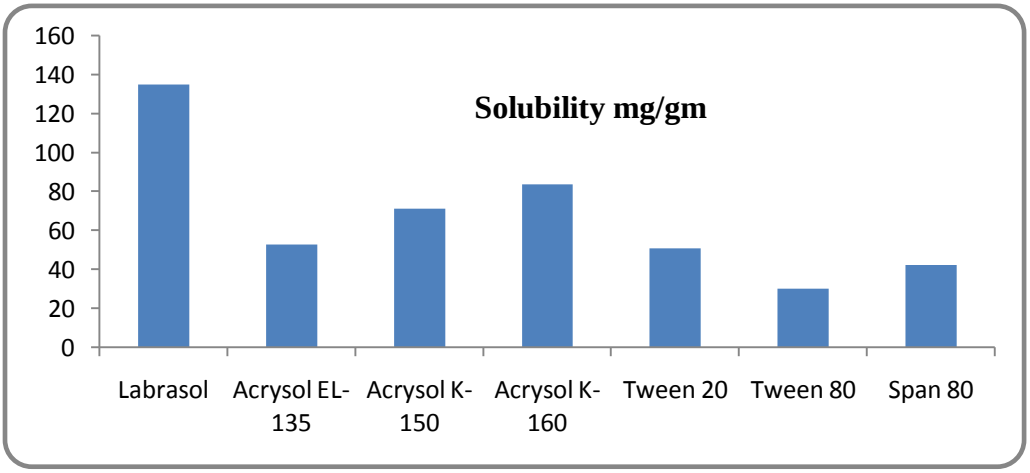


Figure 3.2: Schematic diagram of drug solubility in different surfactant

 International Standard Serial Number (ISSN): 2319-8141

As per solubility data of Carvedilol in different Co-surfactant, maximum amount of drug dissolved in Transcutol CG (125.23 mg/gm).Therefore this Co-surfactant was selected for SMEDDS formulation.

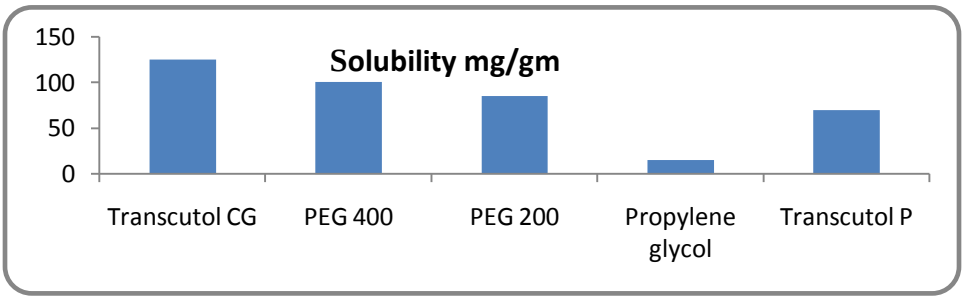


Figure 3.3: Schematic diagram of drug solubility in different Co-surfactant

3.2 Pseudo-ternary phase diagram

The construction of pseudo ternary phase diagram of 1:1 Smix ratios maximum area covered by particular Smix was selected which indicates that the area covers the maximum numbers of formulation.

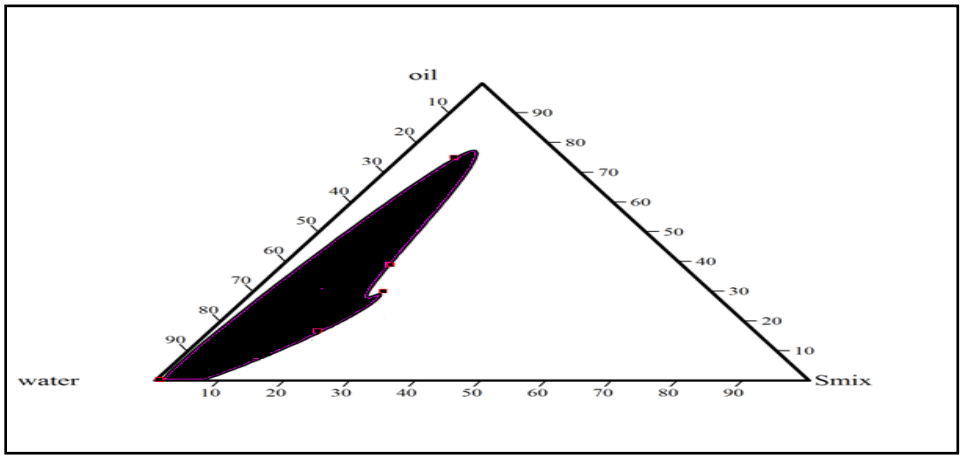


Figure 3.4 Pseudo-ternary phase diagram of S_{mix} ratio 1:1

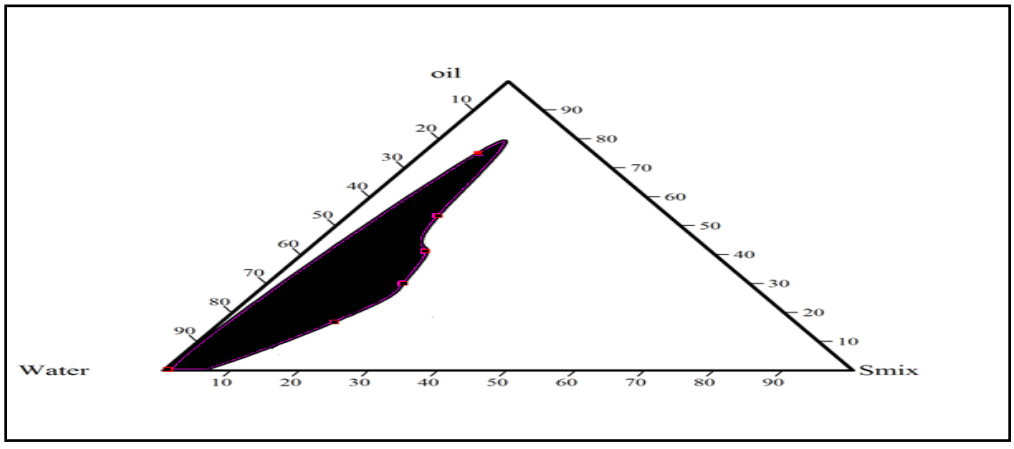


Figure 3.5 Pseudo-ternary phase diagram of S_{mix} ratio 1.5:1

 International Standard Serial Number (ISSN): 2319-8141

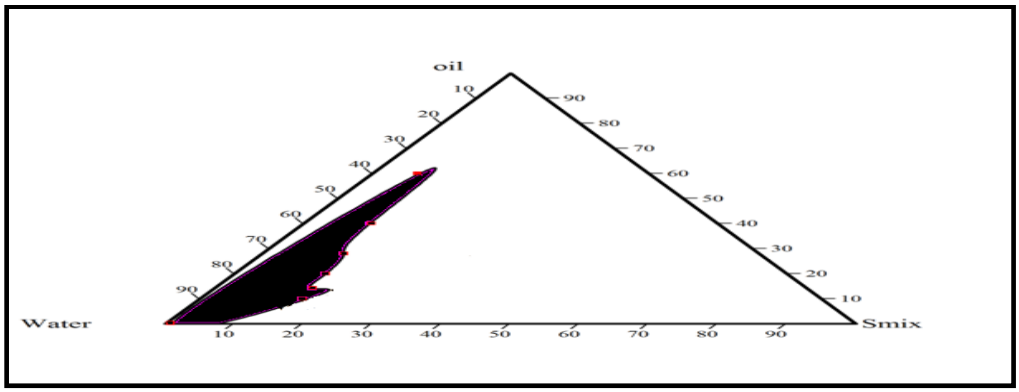


Figure 3.6 Pseudo-ternary phase diagram of S_{mix} ratio 2:1

3.3 Visual assessments

Table: 3.2 Visual assessments of various formulations

Formulation Code	Grade	Formulation Code	Grade
S_1	I	S_4	I
S_2	I	S_5	I
S_3	I	S_6	II

From the table 5.10 data, it was clearly indicate that all the formulations were grade I formulation except S_6 formulation as per dispersibility grade, that means Rapid forming microemulsion (<1 min) which is clear or slightly bluish in appearance.

3.4. Robustness to dilution

Robustness to dilution was performed diluted with excess of water, standard phosphate buffer pH 6.8 and 0.1N HCl 900ml and was stored for 12 hours and result were shown in table 5.16

Table: 3.3 Robustness to dilution

Vehicles	S_1	S_2	S_3	S_4	S_5	S_6
Distilled water	√	√	√	√	√	√
0.1N HCl	√	√	√	√	√	√
Phosphate buffer pH 6.8	√	√	√	√	√	√

Where √ Stable preparation.

As per the above data, all the formulations were stable in all the vehicle and no precipitation or phase separation was found.

3.5 Transmission test

%Transmittance was measured by directly taking the absorbance of the diluted SMEDDS. As per the data of table 5.11, highest %transmittance was reported in S_2 formulation(99.6%). A value of % transmittance

closer to 100% signifying that all of the formulations were clear and transparent. Besides signifying clarity of the formulation, a percentage transmittance closer to 100% also indicates that the size of the globules in the formulation is in the nanometer range. This in turn indicates that the drug in the formulation has a large surface area for release.

3.6 Determination of self-emulsification time

The efficiency of self-emulsification could be estimated primarily by determining the rate of emulsification which is an important index for the assessment of the efficiency of emulsification that is the SMEDDS should disperse completely and quickly when subjected to aqueous dilution under mild agitation.

As per the table 5.12 data, The emulsification time of these formulations were in the range of 15-24sec.

3.7 Drug content

Difference in composition the drug content of formulations S_1 and S_6 was found in range of 96.4-99.8%.

3.8 zeta potential

Formulation S_1, S_2, S_3, S_4 and S_6 have a zeta potential value -12.1, -11.7, -36.8, -13.6, -10.7, -14.5 respectively. This lies in ideal limit mentioned.

3.9 droplet size and Polidispersibility Index

As per the data depicted in above table, droplet size was found to be in range from 18.05-215.3 nm indicating all the particles were in the nanometer range. The smallest particles are observed for formulation S_2, S_4, S_5 and largest particles were obtained for formulation S_1, S_3, and S_6.

The data (table 5.15) showed that formulation S_2 and S_4 have PDI less than 0.3 while in opposite to that S_1, S_3, S_5 and S_6 have greater than 0.3.

It was concluded from above discussion that S_2 formulation exhibited good droplet size, PDI and zeta potential

Table: 3.4 Droplet size, PDI, Zeta potential of S_1 to S_6 Formulation

Formulation code	Average droplet size (water)	PDI	Zeta potential
S_1	159 29nm	0.376	-12.1 mV
S_2	18.3nm	0.239	-11.7 mV
S_3	215.3nm	0.454	-36.8 mV
S_4	50.12nm	0.106	-13.6 mV
S_5	67.3nm	0.346	-10.7 mV
S_6	135.2nm	0.312	-14.5 mV

 International Standard Serial Number (ISSN): 2319-8141

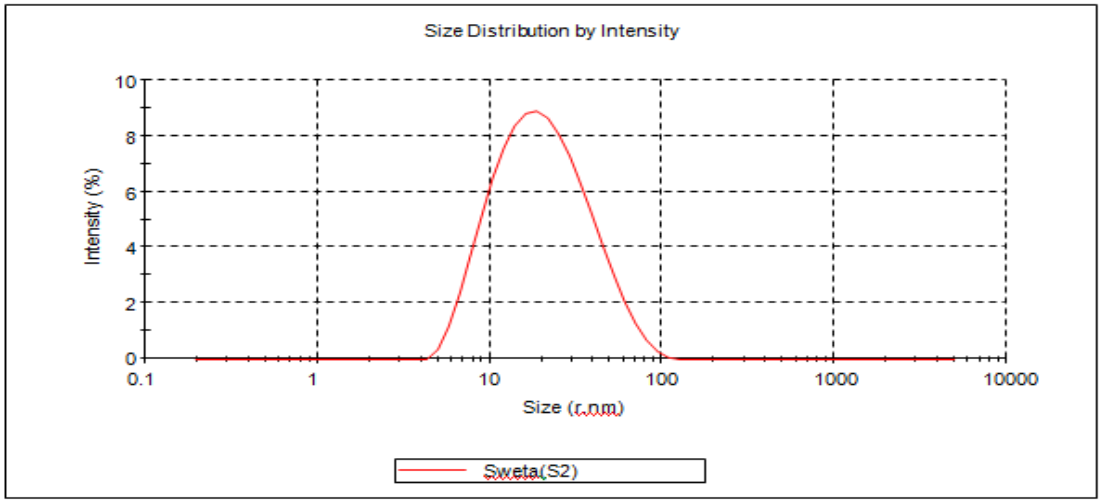


Figure: 3.7 Droplet size Distribution of Batch S_2

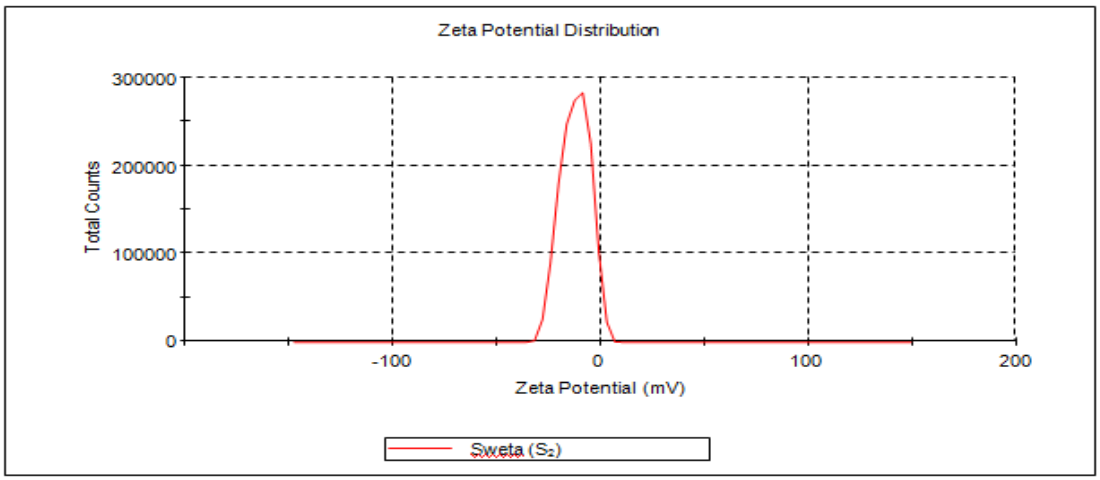


Figure: 3.8 Zeta Potential Determination of Batch S_2

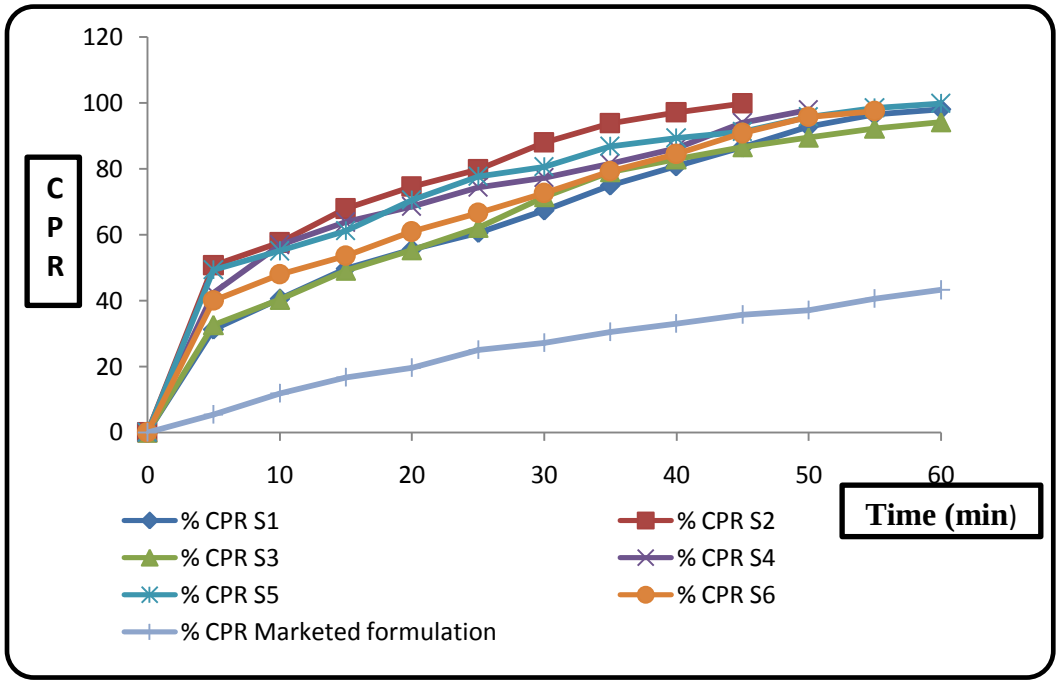
Table 3.5 Emulsification time, % drug content, % transmittance of S_1 to S_6 formulation

Formulation code	Emulsification time (sec)	% Drug content	% transmittance
S_1	18±1	96.4± 0.24	98.3±0.21
S_2	14±1	99.8±0.04	99.6±0.11
S_3	24±1	97.3±0.47	98.2±0.18
S_4	16±1	99.4±0.12	99.2±0.29
S_5	22±0.8	96.9±0.09	99.0±0.06
S_6	19±0.81	98.7±0.16	98.0±0.04

*Values are expressed as mean ± S.D, n=3

 International Standard Serial Number (ISSN): 2319-8141

3.10 In- vitro drug release study



As per data of in-vitro drug release study, it was concluded that S_2 formulation give drug release in 99.85% in 45 min. All the formulations of Carvedilol SMEDDS was compared with the marketed formulation of Carvedilol tablet (12.6 mg). Marketed formulation was give drug release 43.22% while SMEDDS formulation was give more than 90% drug release. So, it was indicate, SMEDDS formulation of Carvedilol was improve the release profile of carvedilol as well as its bioavailability.

4. DISCUSSION

In SEDDS formulation consist of oil, surfactant and co-surfactant. Oil, surfactant and co-surfactant were selected on the basis of solubility and emulsification ability. Capmul MCM NF, Acrysol K160, Transcutol CG were selected on the basis of solubility and emulsification ability for the SEDDS formulation.

Carvedilol was formulated as a SEDDS in an attempt to increase its solubility. An optimized formulation of SEDDS containing carvedilol was developed through the construction of pseudo-ternary phase diagram, *in-vitro* dissolution study, particle size analysis and zeta potential and other evaluation study. SEDDS provided significant increase in the solubility compared to a marketed formulation. SEDDS appeared to be an interesting approach to improve problems associated with oral delivery of carvedilol. Carvedilol SEDDS formulation was superior to marketed formulation with respect to *in-vitro* dissolution profile activity. Thus, SEDDS can be regarded as novel and commercially feasible alternative to current carvedilol formulations.

5. REFFERENCES:

1. Sudheer P, Kumar NM, Puttachari S, Shankar UMS and Thakur RS, "Approaches to development of solid-self micron emulsifying drug delivery system: formulation techniques and dosage forms- A review." *Asian J.Pharm.& Life Sci.***2012**,*2(2)*,214-225
2. Chaus HA, Chopade VV and Chuadhari PD, "Self emulsifying drug delivery system (SEDDS):A review." *Int. J.Pharm.&Chem.Sci.***2013**,*2(1)*,34-44
3. Kang BK, Chon SK and Kim SH, "controlled release of paclitaxel from microemulsion containing PLGA and evaluation of antitumour activity in vitro and in vivo." *Int. J. Pharm.***2004**,*286*,147-156
4. Shukla JB, Koli AR, Ranch KM and Parikh RK, "self emulsifying drug delivery system." *Int. J. Pharm. Sci.***2010**,*1(2)*,13-33
5. Shah NH, Carvajal MT, Patel CI, Infeld MH andMalick AW, "Self emulsifying drug delivery system(SEDDS) with polyglycolized glycerides for improving in vitro dissolution and oral absorption of lopophillic drugs." *Int. J. Pharm.***1994**,*106*,15-23
6. Sunitha R, Satya SD, and Aparna MLV, " Novel Self emulsifying drug delivery system(SEDDS)-An approach to enhance the bioavailability of poorly water soluble drugs." *Int. J. Res. Pharm. & Chem.***2011**,*1(4)*,828-838
7. Mehta K, Borade G, Rasve G and Bandre A, "Self emulsifying drug delivery system (SEDDS) - formulation and evaluation." *Int.J.Pharm. & Bio. Sci.***2011**,*2(4)*,398-412
8. Patil P, Vandana P, Paradkar P., "Formulation of self emulsifying drug delivery system for oral delivery of simvastatin: In vitro and in vivo evaluation", *Acta Pharma*, **2007**, 57,111-122.
9. Patel M., Patel N. and Patel R.; "Formulation and evaluation of self-microemulsifying drug delivery system of lovastatin", *Asian Journal of Pharmaceutical Sciences*, **2010**, 5 (6), 266-275
10. Chae G., Lee J., Kim S., Seo K., Kim M. and Khang G. " Enhancement of the stability of BCNU using self-emulsifying drug delivery systems (SEDDS) and in vitro antitumor activity of self-emulsified BCNU-loaded PLGA wafer", *International Journal of Pharmaceutics*,**2005**; 301: 6–14.
11. Georgakopoulos E, Farah N and Vergnault G., "Oral anhydrous non-ionic microemulsions administered in softgel capsules", B. T. Gattefosse, **1992**; 85: 1120.