https://doi.org/10.33472/AFJBS.6.6.2024.1314-1333

African Journal of Biological Sciences

Journal homepage: http://www.afjbs.com

Research Paper Open Access

Formulation, Characterization and Evaluation of Anti-Oxidant Action of Mangiferin Nanoemulsion

Nilatmaj Singh, Neelam Singh and Surya Prakash Gupta*

Rajiv Gandhi Institute of Pharmacy
Faculty of Pharmaceutical Science & Technology
AKS University, Satna-485001 (MP)-India
***Corresponding Author:**Dr. Surya Prakash Gupta
Email ID: suryatony@yahoo.co.in
Contact No: 07354967776

Article History

Volume 6,Issue 6, 2024
Received:20 Feb 2024
Accepted : 28 Mar 2024
doi: 10.33472/AFJBS.6.6.2024.1314-1333

Abstract

The present investigation was undertaken with an objective to prepare the SNEDDS of mangiferin in order to improve the bioavailability and assess the antioxidant activity of the SNEDSS. Emulsification studies showed that Span 60 was able to produce clear microemulsion with oleic acid upon dilution, and hence, it was employed as the surfactant in further studies. PEG400 was used as the cosolvent for the formulation of SNEDDS. In order to identify the self-emulsifying regions and to optimize the percentages of different liquid SNEDDS components, a ternary phase diagram was constructed in the absence of mangiferin. The results revealed that span 60 and PEG400 1:1 (F4-F6) and 2:1 (F12-F15) exhibited largest nanoemulsion area and shortest emulsification time (less than 1 min). A fixed mangiferin concentration of 5% w/w was selected to be loaded in all self-emulsifying formulations. The prepared formulations were kept in closed containers and tested for thermodynamic stability. The *in vitro* release studies revealed the drug release profiles for the SNEDDS. All the formulations exhibited quick drug release characteristics and almost complete drug release in 15-45 minutes. In contrast, the pure drug exhibited only a maximum of 42.722% release in 60 min duration.

Keywords

Nanoemulsion, mangiferin, antioxidant, ternary phase diagram, release

Introduction

The majority of drugs are hydrophobic (lipophilic) in nature, thus leads to low solubility and bioavailability problems[1,2]. Hydrophobic drug has low oral bioavailability, uncertain absorption profiles, dose variations, wide intra and inter-subject variabilities, and increased chances of food effect. Thus, these drugs express poor therapeutic efficacy[3,4]. Nanoemulsion based drug delivery systems are promising tool for improving the bioavailability of hydrophobic drugs. Nanoemulsion drug delivery systems are effective in solubilizing active lipophilic compounds, and therefore have several applications. Nanoemulsion drug delivery system has shown potential for effective systemic delivery of active components, such as food ingredients and lipophilic drugs, via oral, parenteral, ocular, and topical routes[5]. O/W vitamin nanoemulsions and nutraceuticals facilitate solubilization of these hydrophobic bioactive food components in GIT, therefore increasing bioavailability[6].

Several highly potent hydrophobic drugs have been formulated as nanoemulsion or self-nanoemulsifying drug delivery systems for improving bioavailability and stability of the incorporated drugs[8-13].

Mangiferin is a phytophenolic component isolated from *Mangifera indica* and is known to possess several pharmacological actions like cardioprotection, antimicrobial, anti-inflammatory etc[14]. The low solubility and low permeability of Mangiferin make the clinical use of the molecule difficult. Therefore, there is a need to develop a formulation to improve permeability and bioavailability of Mangiferin so as to reduce the multiple dosing rate and increase the efficacy and patient compliance through the nano-technique based formulation.

Material and Methods

Solubility study in oil and other excipients

The solubility of mangiferin in different oils, surfactants and co-surfactants was determined according to the method of Date and Nagarsenker[15]. In this method, an excess amount of the drug was mixed with fixed amounts of the oil (castor oil, sesame oil, coconut oil, peanut oil, eucalyptus oil, oleic acid, cinnamon oil), surfactants (Tween 80, Tween 20, Span 20, Span 60) and cosurfactants (PEG 400, Propylene glycol, ethanol, butanol) and the mixtures were shaken for 48 hours at 25°C to attain equilibrium. The samples were then centrifuged to remove the undissolved drug, filtered through a 0.45 µm membrane filter, and the supernatant was suitably diluted before spectrophotometric analysis at 257 nm using UV-visible spectrophotometer to determine the amount of the drug dissolved in each excipient.

Surfactant and oil miscibility

The oil and surfactant in the ratio of 1:1 were shaken at 40°C in 3 ml transparent glass vials. The miscibility was monitored optically and considered to be good when the mixture was transparent.

Screening for Emulsifying ability

Surfactant screening

The emulsification ability of different surfactants was evaluated by mixing the surfactant with the selected oily phase in a 1:1 weight ratio. The mixtures were vortex mixed and diluted up to 200 fold dilution. The ease of formation of an emulsion was assessed by observing the number of inversion of the volumetric flask required to obtain a uniform emulsion. The resulting emulsion was also examined visually for relative turbidity according to different grading systems (Grades A – E) described by Khoo et al[16] that depict the spontaneity and appearance of the nanoemulsion formed upon dilution.

Co-surfactant Screening

The ability of co-surfactants (or co-solvents) to improve the emulsification ability of surfactants was also evaluated according to the method of Date and Nagarsenker[15]. Mixtures of the selected oily phase, surfactants and co-surfactants (or co-solvents) were mixed at a ratio of 3:2:1, respectively, and then diluted with distilled water for 200 fold dilution. The appearance and the ease of formation of microemulsion were assessed as described above for screening of surfactants.

Construction of ternary phase diagrams

Based on the solubility of mangiferin, cinnamon oil was chosen as the oil phase. Span 60 was used as the surfactant and PEG 400 was employed as the co-surfactant. Distilled water was used as the aqueous phase for development of these phase diagrams. The surfactant and co-surfactant (Smix) in were mixed in different weight ratios (1:1 **(Smix 1)**, 2:1 **(Smix 2)**, 3:1 **(Smix 3)**) so that the concentration of surfactant increases with respect to co-surfactant.

The oil phase and each Smix were blended thoroughly in 9 different weight ratios (9:1, 8:2, 7:3, 6:4, 5:5, 4:6, 3:7, 2:8, 1:9). From these each ratio, 0.1 ml of mixtures was transferred to separate glass beakers. To these contents, 100 ml distilled water was added gently stirrer using a magnetic stirrer at 37°C.The resulted emulsions were examined for clarity, phase separation, and coalescence of oil droplets on standing for 2 h. In plotting ternary phase diagram, one axis represents the oil phase, the second represents the Smix and the third represents the aqueous phase. The phase diagram was constructed to identify the nanoemulsifying region, using oil and Smix ratios which form ‘good’ emulsions upon dilution with purified water.

Table 1 Composition for construction of ternary phase diagram (%w/w)

Formulation	Oil	Smix 1	Smix 2	Smix 3

F1	9	1	-	-
F2	8	2	-	-
F3	7	3	-	-
F4	6	4	-	-
F5	5	5	-	-
F6	4	6	-	-
F7	3	7	-	-
F8	2	8	-	-
F9	1	9	-	-
F10	9	-	1	-
F11	8	-	2	-
F12	7	-	3	-
F13	6	-	4	-
F14	5	-	5	-
F15	4	-	6	-
F16	3	-	7	-
F17	2	-	8	-
F18	1	-	9	-
F19	9	-	-	1
F20	8	-	-	2
F21	7	-	-	3
F22	6	-	-	4
F23	5	-	-	5
F24	4	-	-	6

F25	3	-	-	7
F26	2	-	-	8
F27	1	-	-	9

Preparation of mangiferin-loaded self-nanoemulsifying formulations (SNEDDs)

Mangiferin was added to the optimized blank ternary systems at a drug loading concentration of 5% w/w. Final mixtures were mixed and shaken for 24 hours at 25°C in a shaking water bath to ensure complete solubilization.

Table 2 Composition of optimized ternary systems for SNEDDs

Formulation	Oil %w/w	Surfactant %w/w	Cosurfactant %w/w	Smix ratio
F4	60	20	20	1:1
F5	50	25	25	1:1
F6	40	30	30	1:1
F12	70	20	10	2:1
F13	60	26.6	13.3	2:1
F14	40	45	15	2:1
F15	30	52.5	17.5	2:1
F23	50	37.5	12.5	3:1

Evaluation of optimized SNEDDS formulation

Thermodynamic stability studies and cloud point

Stability of the optimized SNEDDS formulation was evaluated at different stress conditions such as heating cooling cycles (4°C and 40°C) and freeze thaw cycles (-21°C and +25°C) along with storage at specified temperature for 48 h. In order to carry out centrifugation stress study, 1 mL of the formulation was diluted to 100 mL with distilled water and centrifuged at 10000 g for 20 min and visually observed for any phase separation[17]. In order to determine cloud point temperature, 10 mL of diluted SNEDDS formulation were gradually heated on a water bath and observed for cloudiness using thermometer. The temperature at which cloudiness appeared was denoted as cloud point.

Measurement of particle size

The particle size and polydispersity index of the SNEDDS was obtained using a dynamic light scattering particle size analyzer. The SNEDDS were dispersed in purified water and placed in the path of the laser beam. The particle size and polydispersity was recorded[18].

Measurement of zeta potential

The zeta potential of selected formulation was determined using Zetasizer. Samples were properly diluted with deionized water (1:200) and filtered through a 0.45 µm membrane filter before measurement.

Determination of drug content of mangiferin-loaded SNEDDS

An accurately weighed amount of the resulting drug-loaded SNEDDS formulation was dispersed in a suitable quantity of methanol and shaken thoroughly to ensure release and dissolution of the drug in methanol. The samples were centrifuged at 3000 rpm for 15 minutes and the supernatant was filtered through a 0.45 µm membrane filter and the filtrate

was assayed spectrophotometrically for the drug at a wavelength of 257 nm. The drug content in each sample was calculated as milligrams of the drug per gram of the product using the following equation:

$$drug\ content = \frac{drug\ content\ in\ the\ weight\ taken\ from\ solid\ \ SNEDDS}{weight\ of\ the\ solid\ SNEDDS\ taken}$$

The experiments were repeated in triplicate for each produced batch and then the results were averaged ± standard deviation.

***In vitro* release study**

The *in vitro* release studies of different mangiferin SNEDDS formulations were carried out in dissolution apparatus II (Paddle method) by dialysis bag method. The dissolution medium composed of 900 ml phosphate buffer pH 7.2 maintained at 37 ± 0.5°C and the rotational speed was adjusted at 50 rpm. Phosphate buffer pH 7.2 was prepared by mixing 50 ml of 0.2M potassium dihydrogen orthophosphate with 35 ml of 0.2M sodium hydroxide and diluting to 200 ml with water. An amount of SNEDDS formulation equivalent to 25 mg of mangiferin was filled in dialysis membrane and used for dissolution studies. Samples were withdrawn at predetermined time intervals. An equal volume of fresh dissolution medium maintained at the same temperature was added to keep constant volume during dissolution study. The collected samples were filtered through 0.45 µm syringe filter, suitably diluted using methanol and then assayed for the content of mangiferin by UV spectrophotometry at 257 nm.

Antioxidant Study

The free radical scavenging activity of the test solution was measured in terms of hydrogen donating or radical scavenging ability using the stable free radical DPPH.

Determination of DPPH radicals scavenging activity was performed by the previously reported method[19]. Separately, 1mM solution of DPPH and test solution (50-250 µg/mL) were prepared in ethanol. 1.5ml of the test solution was added to 1.5 ml of DPPH solution. The absorbance was measured at 517 nm against the corresponding blank solution which was prepared using 3 mL ethanol. The control sample used was 3 mL of DPPH. The assay was performed in triplicates. Percentage inhibition of free radical DPPH was calculated based on control reading by following equation.

$$\text{DPPH scavenged (\%)} = \frac{(A_{con} - A_{test})}{A_{con}} \times 100$$

A_{con} - is the absorbance of the control reaction

A_{test} - is the absorbance in the presence of the test solution.

Results and Discussion

FTIR of mangiferin

The IR spectrum of the drug sample of mangiferin was obtained and the stretching and bending vibrations of OH, C-H and C-O were observed (Figure 1).

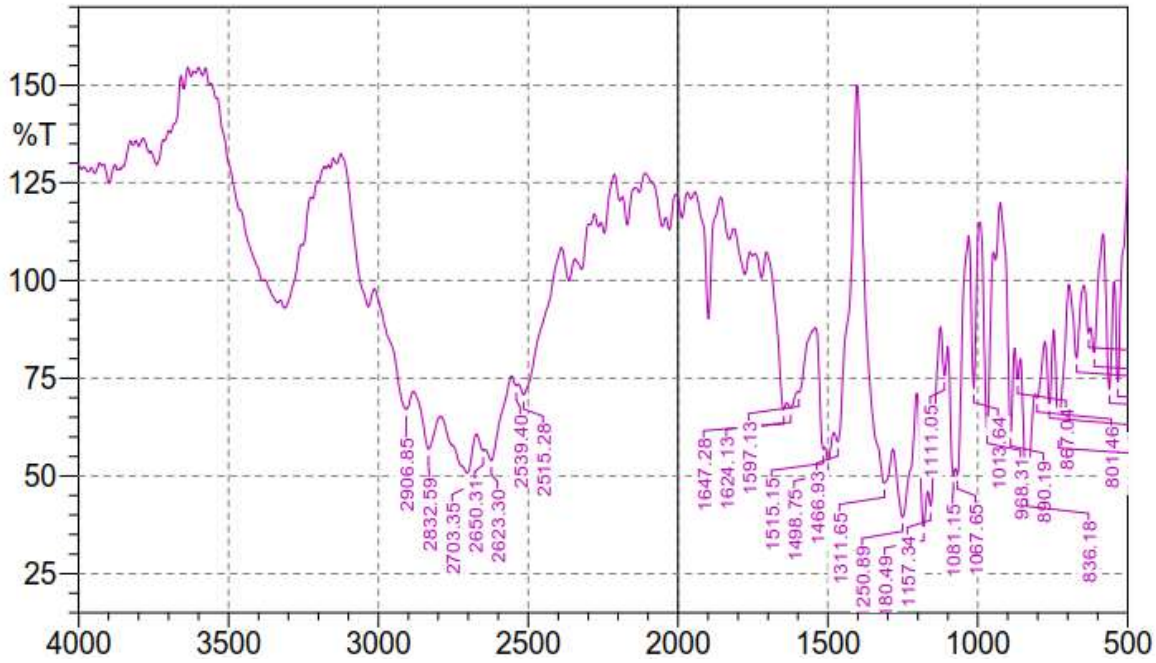

Figure 1 FTIR spectra of mangiferin

Calibration curve of mangiferin

The absorption maximum of mangiferin in methanol was found to be 257 nm and the calibration curve was prepared for a range of 10-50 µg/mL (Figure 2).

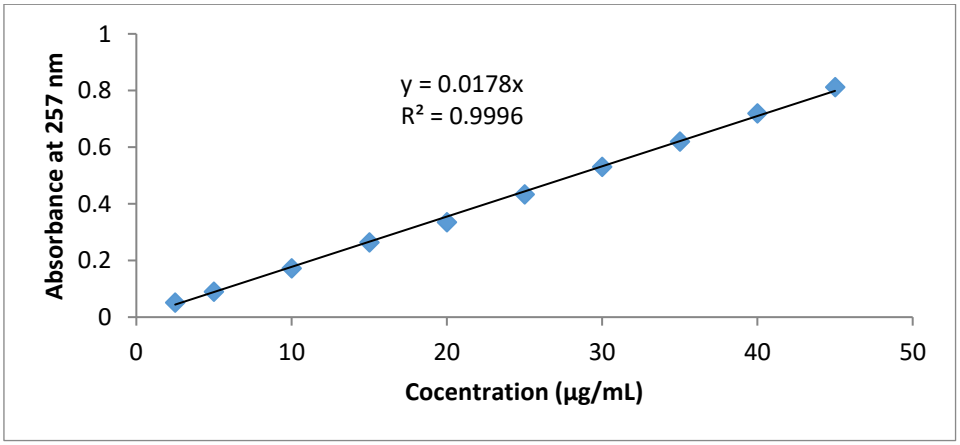

Figure 2 Standard curve of mangiferin

Solubility Studies

The solubility of simvastatin was determined in oils, surfactants, co-surfactants, mixture of oils and mixture of surfactants (Figure 3).

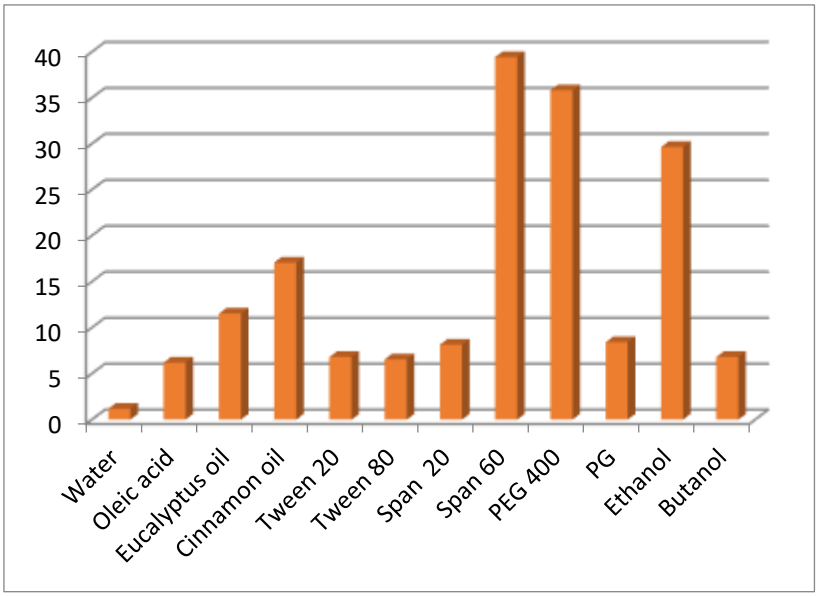

Figure 3 Solubility of mangiferin

Among the tested oils, mangiferin exhibited the highest solubility in cinnamon oil compared to all other oils. To obtain a clear micro-emulsion proper selection of oil, surfactant, co-surfactant/cosolvent and oil to surfactant/co-surfactant ratio is significant. Cinnamon oil was selected as the oil phase form preparing the micro-emulsion. The highest solubility was exhibited by Span 60 and it has an HLB value of 4.7.

Selection of surfactant and co-surfactant

Selection of surfactants should be based on its emulsification efficiency for the selected oil more than its solubilizing potential for the drug[20]. Therefore, the miscibility of the above surfactants with the selected oil (cinnamon oil) at a 1:1 weight ratio was investigated according to the method reported by Balakrishnan[21] and Date and Nagarsenker[15]. Emulsification studies showed that Span 60 was able to produce clear microemulsion with cinnamon oil upon dilution, and hence, it was employed as the surfactant in further studies.

The use of a single surfactant may not be enough to achieve a transient negative interfacial energy or a fluid interfacial film. Hence, addition of a co-surfactant/cosolvent may provide sufficient flexibility to the interfacial film so that various curvatures can be available to form microemulsions over a wide range of composition. The co-surfactant and co-solvents used were equivalent in improving emulsification ability of surfactants as demonstrated by grades A and B produced upon dilution with distilled water. Hence blends of span 60 and PEG 400 were used for the formulation of the microemulsions. The appropriate amounts of the selected oil, surfactants and co-solvent were determined by constructing phase diagrams.

Construction of ternary phase diagram

In order to identify the self-emulsifying regions and to optimize the percentages of different liquid SNEDDS components, a ternary phase diagram was constructed in the absence of mangiferin.

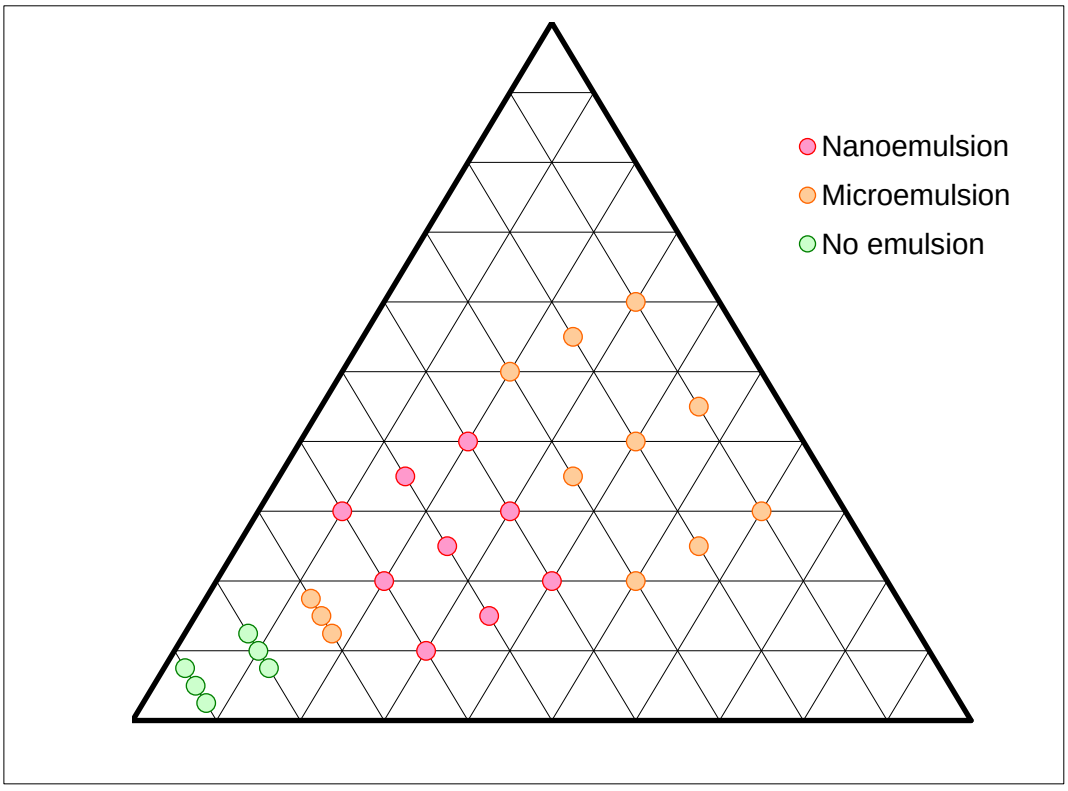

Figure 4a Ternary phase diagram for Smix 1-water-cinnamon oil

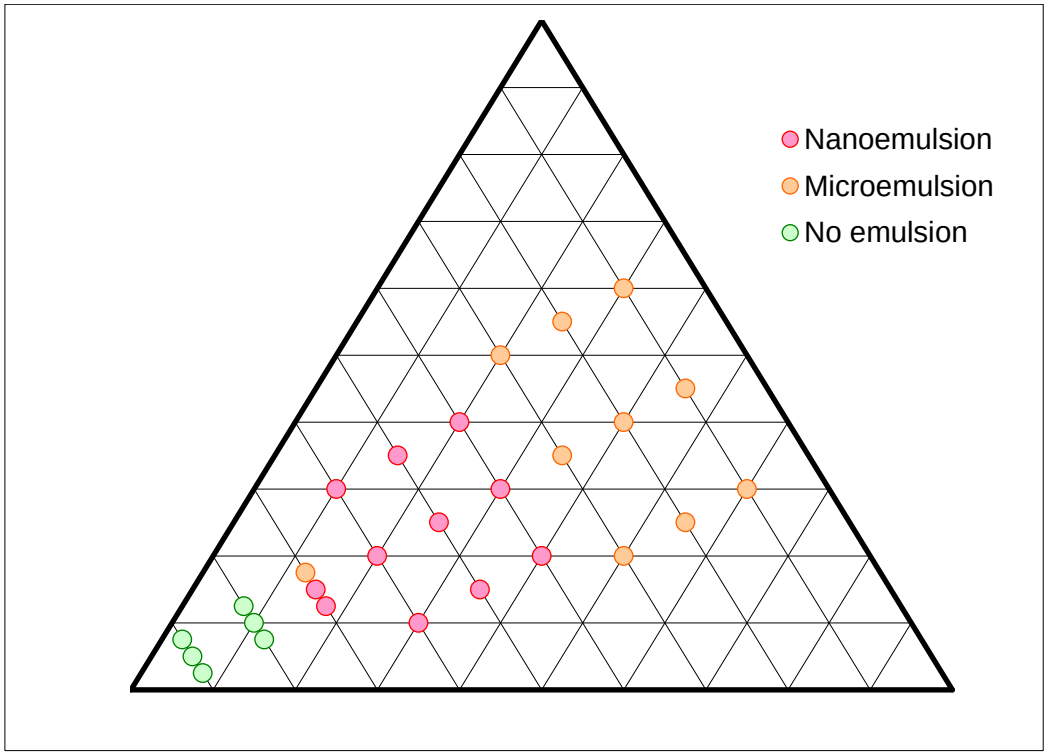

Figure 4b Ternary phase diagram for Smix 2-water-cinnamon oil

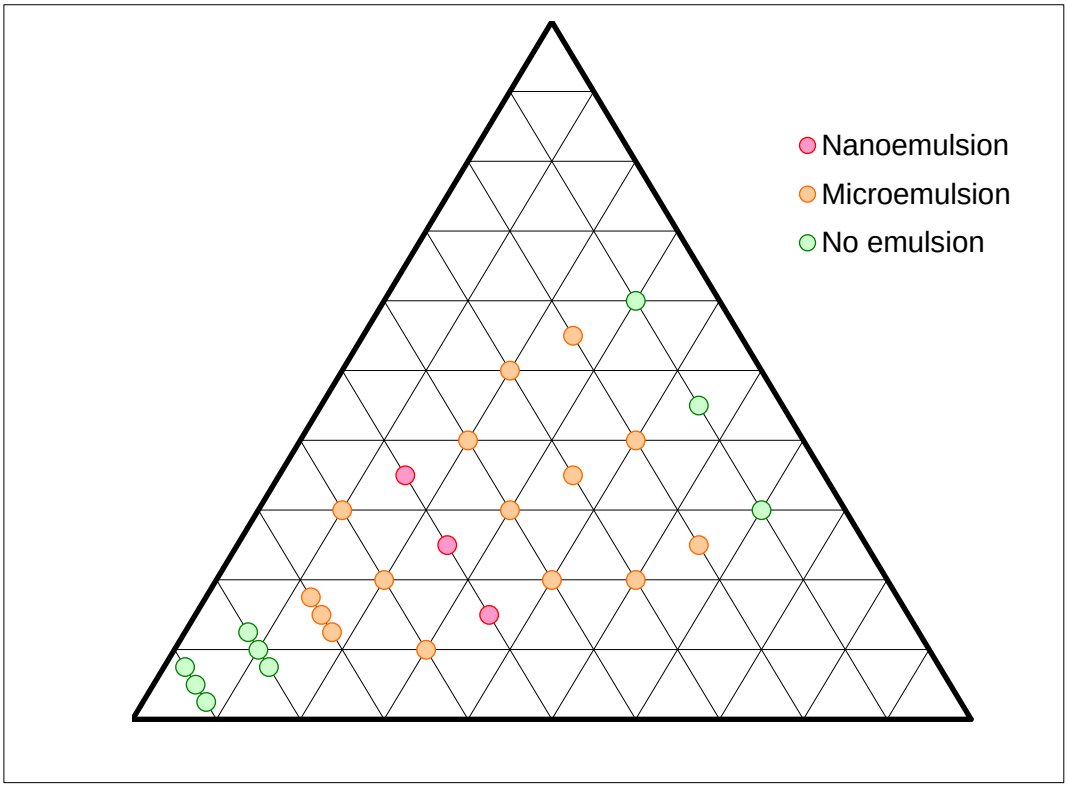

Figure 4c Ternary phase diagram for Smix 3-water-cinnamon oil

Different batches of SNEDDS were formulated and visually observed for their self-emulsifying properties. The ternary phases were judged as nanoemulsions, microemulsion and no emulsion formation on the basis of their turbidity measurements and visual observations for transparency. The concentration of components was expressed as percent volume/volume (% v/v) in ternary phase diagram. The results revealed that span 60 and PEG 400 used in ratios of 1:1 (F4-F6) and 2:1 (F12-F15) exhibited largest nanoemulsion area and shortest emulsification time (less than 1 min). In the Smix ratio 3:1, only F23 led to the formation of nanoemulsions. It was observed that with increase in the ratio of the PEG400, spontaneity of the self-emulsification process got increased. The transparent emulsions (F4, F5, F6, F12, F13, F14, F15 and F23) were visually evaluated for clarity and stability after 48h at room conditions. All tested emulsions remained clear transparent even at the end of 48h. Hence, these ternary phases were selected for mangiferin loaded SNEDDs.

Mangiferin -loaded self-microemulsifying formulations (SNEDSS)

The ternary phase diagrams revealed the optimum concentration of the oil and the surfactant mix that could be used for the formulation of mangiferin loaded SNEDDs. A fixed simvastatin concentration of 5% w/w was selected to be loaded in all self-emulsifying formulations. It was expected to provide spontaneous emulsification of SNEDDS with a low tendency of drug precipitation upon aqueous dilution. Also, using fixed concentration of mangiferin in all formulations was proposed to exclude the effect of varying the drug concentration on the self-emulsifying efficiency of the systems.

Thermodynamic stability and cloud point determination

The prepared formulations were kept in closed containers and tested for thermodynamic stability. Thermodynamic stability studies were carried out to determine the effects of

temperature variation and centrifugation on precipitation or phase separation of the formulated SNEDDS.

All the formulations passed the thermodynamic stability studies without any signs of phase separation and precipitation during alternative temperature cycles (4°C and 40°C), freeze thaw cycles (-21°C and +25°C) and centrifugation at 10,000 g indicating good stability of formulations and their emulsions. Determination of cloud point is an essential parameter for the selection of a stable SNEDDS particularly when composed with non-ionic surfactants. "The cloud point temperature (lower consolute temperature) indicates the temperature at which the transparent monophasic system was transformed into cloudy biphasic system as dehydrated surfactant molecules associated together as precipitate, which can affect the formulation adversely. It is recommended that the cloud point for SNEDDS should be higher than body temperature (37°C), which will avoid phase separation occurring in the gastrointestinal tract. The cloud point temperature of the tested SNEDDS was found to be in the range of 90.73-96.32°C (Table 6.7). Thus, it can be inferred that the developed formulation was stable and do not require a precise storage temperature and it develops a stable emulsion upon administration at physiological temperature in vivo.

Droplet Size, Polydispersity and zeta potential of SNEDDs

The mean droplet size and polydispersity index (PDI) determined for different mangiferin-loaded SNEDDS (F7-8, F15-16) are shown in Table 3.

Incorporation of different amount of Smix into mangiferin-loaded SNEDD formulations resulted in significantly different droplet size. Among the tested formulations, SNEDDS formulations prepared with Smix 2 exhibited lower droplet size compared to formulations with Smix 1 and Smix 3.

Table 3: Stability and characterization of SNEDDS

Formulation	Thermodynamic Stability				Surface Characterization		
	Cloud point (°C)	**Centrifugation**	**Cooling/Heating**	**Freeze/Thawing**	**Mean droplet size**	**PDI**	**Zeta potential**
F4	90.73	No phase separation	No Phase inversion	No Phase inversion	512.92 ± 6.07	0.613 ± 0.008	-21.5
F5	91.65	No phase separation	No Phase inversion	No Phase inversion	493.21 ± 5.03	0.719 ± 0.004	-26.8
F6	91.96	No phase separation	No Phase inversion	No Phase inversion	471.03 ± 8.08	0.763 ± 0.003	-28.9
F12	94.58	No phase separation	No Phase inversion	No Phase inversion	457.32 ± 8.15	0.801 ± 0.008	-27.5
F13	94.79	No phase separation	No Phase inversion	No Phase inversion	449.35 ± 7.05	0.568 ± 0.008	-25.3
F14	95.11	No phase separation	No Phase inversion	No Phase inversion	427.41 ± 8.38	0.418 ± 0.008	-25.9
F15	95.19	No phase separation	No Phase inversion	No Phase inversion	406.57 ± 7.14	0.523 ± 0.008	-27.1
F23	96.32	No phase separation	No Phase inversion	No Phase inversion	461.22 ± 9.15	0.798 ± 0.008	-23.6

It was observed from the results that decreasing the oil content of the formulations resulted in a decrease in the size of formulation droplets.

Self-emulsifying formulations possess a negative charge on the oil droplets due to the presence of anionic groups of free fatty acids contained in their composition; the oil, surfactant and co-surfactant. The obtained high negative values of zeta potential indicate that the tested formulations are less likely to flocculate or aggregate during storage or in biological environment.

In vitro dissolution study

The *in vitro* release studies revealed the drug release profiles for the SNEDDS. All the formulations exhibited quick drug release characteristics and almost complete drug release in 15-45 minutes (Figure 5). In contrast, the pure drug exhibited only a maximum of 42.72% release in 60 min duration.

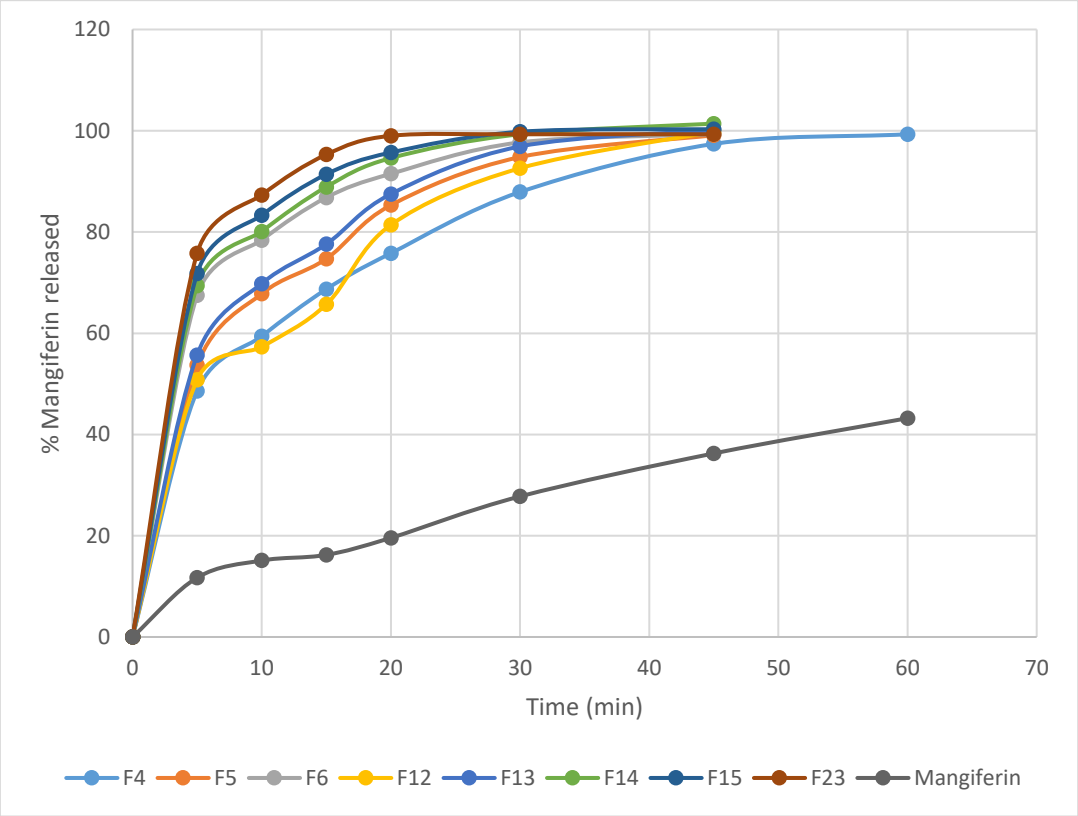

Figure 6.7 *In vitro* release profile of mangiferin from SNEDDS

Mangiferin-loaded liquid SNEDDS formulations (F4, F5, F6, F12, F13, F14, F15 & F23) exhibited optimal release performance. High release profiles of liquid SMEDDS are due to quick formation of o/w nanoemulsions with small droplet size upon exposure to dissolution medium with gentle agitation. In addition, the presence of the drug in a dissolved state in liquid SNEDDS formulations avoids the release rate-limiting step required for crystalline drugs.

Antioxidant Study of Nanoemulsion (F12)

DPPH is stable nitrogen centered free radical that can accept an electron or hydrogen radical to become a stable diamagnetic molecule. DPPH radicals react with suitable reducing agents, then losing colour stoichometrically with the number of electrons consumed, which is measured spectrophotometricallty at 517 nm. The deep purple color of DPPH decreases if the compound exhibits antioxidant action.

Table 4 DPPH radical scavenging potential of SNEDDS

SNEDDS	DPPH Scavenging %	
	SNEDDS at concentration 50µg/mL	Mangiferin at concentration 50µg/mL
F4	41.15 ± 0.521	43.47 ± 0.572
F5	42.17 ± 0.336	-
F6	41.37 ± 0.674	-
F12	42.52 ± 1.033	-
F13	40.28 ± 1.116	-
F14	41.25 ± 0.713	

F15	41.67 ± 0.566	
F23	42.05 ± 0.817	

Results are reported as mean ± SD (n=3)

The SNEDDS at dose of 50µg/mL mangiferin were found to produce protection against formation of DPPH radical almost equal to that exhibited by pure mangiferin solution.

Conclusion

The bioavailability of the lipophilic drugs can be enhanced by formulating them as SNEDDS. From the release behavior witnessed through the present investigation it could be proven that the release and in turn bioavailability of the mangiferin could be doubled by formulating it as SNEDDS. The formulated SNEDDS also exhibited antioxidant action equal to that to pure mangiferin.

Acknowledgements

The authors would like to thank RB Science, Bhopal for providing facilities to perform the release studies.

References

1. Mu, H., HolmR. and Mullertz A. (2013).Lipid-based formulations for oral administration of poorly water-soluble drugs.*Int. J. Pharm*. **453(1)**:215-24.
2. Qian,C. and McClements, D.J. (2011).Formation of nanoemulsions stabilized by model food-grade emulsifiers using high-pressure homogenization: Factors affecting particle size.*Food Hydrocoll*.**25(5)**:1000-8.
3. Chatterjee, B.,Hamed Almurisi, S., Ahmed Mahdi Dukhan, A.,Mandal, U.K.andSengupta, P.(2016).Controversies with self-emulsifying drug delivery system from pharmacokinetic point of view.*Drug Deliv*.**23(9)**:3639-52.

4. Dokania,S. andJoshi, A.K.(2015).Self-microemulsifying drug delivery system (SMEDDS) – challenges and road ahead. *Drug Deliv*. **22(6)**:675-90.
5. Solans, C., Izquierdo, P.,Nolla, J.,Azemar,N. and Garcia-Celma, M.J. (2005).Nano-emulsions.*Curr Opin Colloid Interface Sci*. **10(3-4)**: 102-10.
6. Komaiko, J.S.andMcclements, D.J. (2016).Formation of food-grade nanoemulsions using low-energy preparation methods: A review of available methods. *Compr. Rev. Food Sci. Food Saf.***15(2)**: 331-52.
7. Tiwari, S.B.,Shenoy, D.B. andAmiji,M.M. (2006).Nanoemulsion formulations for improved oral delivery of poorly soluble drugs. *NSTI-Nanotech*. **1**: 475-78.
8. Kawakami, K., Yoshikawa, T.,Hayashi, T.,Nishihara,Y. and Masuda, K.(2002).Microemulsion formulation for enhanced absorption of poorly soluble drugs. *J. Control Release*. **81(1-2)**: 75-82.
9. Sole, I.,Solans, C.,Maestro, A.,Gonzalez, C.andGutierrez, J.M.(2012).Study of nano-emulsion formation by dilution of microemulsions.*J. Colloid Interface Sci.* **376(1)**: 133-39.
10. Montes de Oca-Avalos, J.M.,Candal,R.J. and Herrera,M.L. (2017).Nanoemulsions: stability and physical properties. *Curr. Opin. Food Sci.***16**: 1-6.
11. Anton, N.and Vandamme, T.F. (2009).The universality of low-energy nano-emulsification. *Int. J. Pharm.***377(1-2)**: 142-47.
12. Rai, V.K.,Mishra, N.,Yadav, K.S. andYadav, N.P.(2018).Nanoemulsion as pharmaceutical carrier for dermal and transdermal drug delivery: Formulation development, stability issues, basic considerations and applications. *J. Control Release*. **270**: 203-25.

13. Goncalves,A. Nikmaram, S., Roohinejad*et al*. (2018).Production, properties, and applications of solid self-emulsifying delivery systems (S-SEDS) in the food and pharmaceutical industries. *Colloids Surfaces A.Physicochem. Eng. Asp.***538**: 108-26.
14. https://pubchem.ncbi.nlm.nih.gov/compound/Mangiferin
15. Date,A.A. andNagarsenker, M. (2007).Design and evaluation of self-nanoemulsifying drug delivery systems (SNEDDS) for cefpodoxime proxetil.*Int. J. Pharm.***329**:166-72.
16. Khoo, S.M.,Humberstone, A.J.,Porter, C.J.H.,Edwards,G.A. and Charman, W.N.(1998).Formulation design and bioavailability assessment of lipidic self-emulsifying formulations of halofantrine. *Int. J. Pharm.***167**:155-64.
17. Inugala, S.Eedara, B.B. andSunkavalli, S.(2015).Solid self-nanoemulsifying drug delivery system (S-SNEDDS) of darunavir for improved dissolution and oral bioavailability: in vitro and in vivo evaluation. *Eur. J. Pharm. Sci.***74**: 1-10.
18. Mishra, B.,Pati, J.C.,Mishra,R., andSingh, M.(2023).L-Threonine decorated paclitaxel poly (l-lactide) nanoparticles: formulation, pharmacokinetic and stability study.*J. Popul. Ther. Clin. Pharmacol.***30(16)**: 713-17. DOI: 10.53555/jptcp.v30i16.2570
19. Mishra, N.,Jain,P. andMishra,B. (2017).Derivatization of Gallic Acid with amino acids for accentuation of its antioxidant potential. *J. Pharmacol. Biomed.* **1(3)**: 94-102.
20. Duc Hanh, N.,Mitrevej, A.,Sathirakul, K.,Peungvicha,P. andSinchaipanid, P.(2015).Development of phyllanthin-loaded self-microemulsifying drug delivery system for oral bioavailability enhancement. *Drug Dev.Ind.Pharm.***41**:207-17.
21. Balakrishnan, P.,Lee, B.J.,Oh, D.H.,Kim, J.O.,Hong, M.J.,Jee,J.P. *et al.*(2009).Enhanced oral bioavailability of dexibuprofen by a novel solid Self-emulsifying drug delivery system (SEDDS). Eur.J. Pharm.Biopharm.**72**: 539-45.