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FORMULATION AND EVALUATION OF SELF-NANOEMULSIFYING DRUG DELIVERY SYSTEM FOR RESVERATROL

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ABSTRACT

Resveratrol is a member of the stilbene family emerged as a leading candidate for improving health span through potentially slowing the aging process and preventing chronic diseases. The present investigation deals preformulation, solubility assessment and screening of oils, surfactants and cosurfactants combinations for resveratrol formulation design. Preformulation data is helpful to initiate formulation development of resveratrol molecule. In this research, preformulation studies for resveratrol have been performed to determine factors which influence solubility and stability which, in turn, can be used to assist future formulation development of resveratrol. Solubility assessment and screening of oils, surfactants and cosurfactants combinations for resveratrol was done to enhance its solubility which in turn can enhance bioavailability. Optimized formulation composed of Labrafil M 2125(4.98 w/w), Kolliphor ELP (52.18 w/w), Ethanol (26.01 w/w). The *in vitro* release was found to be higher in formulation containing combinations of these components in comparison to pure resveratrol. Results obtained indicated that HLB value, oil solubility, hydrophobic chain length, greatly influence microemulsion and nanoemulsion. The study demonstrated that solid lipid nanoparticle, self-emulsifying drug delivery system, liposome formulation will be the promising strategies to enhance solubility, stability and in vitro release of resveratrol.

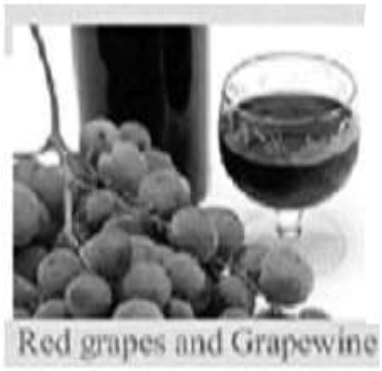
KEYWORDS: resveratrol, solubility, surfactants, nanoemulsion, cosurfactants.

INTRODUCTION

Resveratrol (*trans*-3, 5, 4'-trihydroxystilbene; RESVERATROL) is a naturally occurring polyphenol found in more than 70 plant species including foods (Figure 1). [1]

Table 1: Sources of resveratrol.

Source	Quantity	Resveratrol (mg)
Peanuts	146 g	0.01 – 0.26
Peanut butter	258 g	0.04 – 0.13
Red grapes	160 g	0.24 – 1.25
Cocoa powder	200 g	0.28 – 0.46



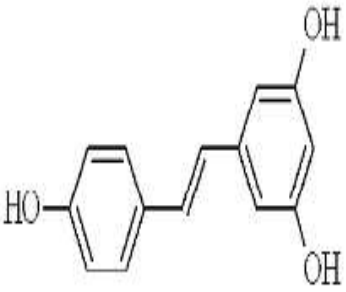


Fig 1: Chemical Structure of Trans-Resveratrol.

In grapes, *trans*-resveratrol is a phytoalexin produced against the growth of fungal pathogens such as Botrytis cinereal.[2,3] In grapes, resveratrol is found primarily in the skin,[4] and, in muscadine grapes, also in the seeds. [5] It is also found in pinus strobus, the eastern white pine. The levels of resveratrol found in food varies greatly (Table 1). Red wine contains between 0.2 and 5.8 mg/ml.[6] Resveratrol is a member of the stilbene family emerged as a leading candidate for improving health span through potentially slowing the aging process and preventing chronic diseases. It has been found to potentially exhibit anticancer, antiangiogenic, immunomodulatory and cardio protective activities, in addition to its usefulness in the treatment of neurodegenerative disease, diabetes and cardiac ailments.[3] Its biological activities have been shown to depend on its structural determinants including the number and position of carboxyl groups, intramolecular hydrogen bonding, stereoisomer, and the presence of double bond. Trans-stilbene compounds which possess ortho-diphenoxyl or para-diphenoxyl functionalities having a 4′-hydroxyl group and double bond show high chemo preventive activity.[7,8] Researchers have investigated the structure-activity relationship by changing the number and the position of hydroxyl groups.[9]

Various research problems have been associated with resveratrol. Resveratrol has shown to be quite unstable. Any preparation of resveratrol typically contains the two common forms of the molecule: the cis- and the trans-resveratrol variants, with the cis-variant being more stable than the trans-variant. This results in the rapid degradation of trans-resveratrol into cis-resveratrol. Unfortunately, only the less stable trans variant is biologically active. This degradation is rapid and well documented: that there is a significant decline in the content of trans-resveratrol in several products during the products shelf-life - up to 55% less than the stated amount.[10] Resveratrol is characterized by low bioavailability, and only a small fraction of it reaches the blood. For example, Brown et al reported Cmax levels of resveratrol following administration of 500-mg daily doses for a minimum of 21 days to be only about 43.8 micrograms per liter of blood.[11] The oral absorption of resveratrol in humans is about 75% and is thought to occur mainly by trans epithelial diffusion. Extensive metabolism in the intestine and liver results in an oral bioavailability considerably less than 1%.[11,12] Resveratrol has wide solubility ranging from 0.05 mg/mL in water to 374 mg/mL in polyethylene glycol 400 (PEG-400). It has extremely low solubility in water Resveratrol is relatively stable above pH 6 and has maximum degradation at pH 9.[13,14] Resveratrol is more soluble in alcohol and PEG-400 and stable in acidic pH.[11]

The present investigation deals solubility assessment and screening of oils, surfactants and cosurfactants combinations for resveratrol formulation design. Preformulation data will be helpful to initiate formulation development of resveratrol molecule. Solubility assessment and screening of oils, surfactants and cosurfactants combinations for resveratrol. It will enhance its solubility which in turn can enhance bioavailability.

MATERIALS

Resveratrol purchased from "Avans Cure Pharmaceutical Company, India" , Ethanol Absolute purchased from "Changshu Yanguan Chemicals, China", Methanol, purchased from "Thermo Fischer Scientific India Pvt. Ltd., Mumbai", Disodium Hydrogen orthophosphate, Potassium Dihydrogen orthophosphate, Sodium hydroxide bought from "Thomas baker (chemicals) Pvt. Ltd. Mumbai, India", n-Octanol, Ethyl Oleate, Oleic acid, PEG200, PEG400 were purchased from SDFCL, Mumbai, Myritol purchased from " BASF, Mumbai, India", Tween20, Tween60, Tween80 purchased from " Molychem, Mumbai, India", Kolliphor RH 40, Kolliphor HS 15, Kolliphor ELP, PEG 300 purchased from "BASF, Germany", Iso Propyl Alcohol, Glycerol were bought from "Avantor Performance Materials Limited,

Haryana, India", Propylene Glycol obtained from "Qualikems Fine Pvt. Ltd., Vadodara,India", Labrafil M 2125 was obtained from "Gattefosse, Germany".

METHODS

Solubility of Resveratrol in Lipids, Surfactant, Co Surfactants

Shake flask method.[15,16] was used for solubility studies of resveratrol in various lipids (Labrafil M 2125, Ethyl Oleate, Oleic acid), surfactants (Kolliphor HS15, Kolliphor ELP, Kolliphor RH 40 and Tween80) and co surfactants (Ethanol, PEG 200, PEG400, PEG 300, n-butanol and Propylene Glycol). An excess amount of resveratrol was added in 2ml of lipophilic in stoppered vials and mixed with vortex shaker (Remi CM 101, International Mumbai, and India). These vials were then kept at 25°C for 24 hours in water bath shaker (Nirmal International, Delhi, India). The resulting samples were centrifuged at 2000rpm for 15 mins (Remi, International Mumbai, and India). The supernatant was filtered through 0.22µm filter. The concentration of resveratrol was then quantified using HPLC (Shimadzu, Kyoto, Japan) instrument equipped with two LC-10 ATVP pumps, SPD-10AVP UV-vis detector.

Screening of Surfactants and Co surfactants

Screening was done by number of flask inversions technique.[17,18] During screening oil and surfactant were taken in ratio 1:1. 150mg of surfactant was added to 150mg of oily phase respectively and then this mixture was heated at 50ºC for homogenization of the components. Whereas during cosurfactant screening oil: surfactant: co-surfactant was taken in the ratios of 3:2:1, the best possible combination as reported in earlier literature.[17] Then from each mixture prepared 100 mg of sample was withdrawn and diluted to 100ml in a volumetric flask. The ease of emulsification was judged by the number of flask inversions required to yield homogeneous emulsion. The emulsions could stand for 24 hrs. and then % transmittance was evaluated at 629 nm by using UV spectrophotometer.

Optimization of oil, surfactant and cosurfactant combinations

The surfactant and co-surfactant were varied in mass ratios 1:1, 1:2, 2:1 for each combination selected by screening. The different concentration ratios of oil: Smix (surfactant and cosurfactant) as per pseudo ternary phase diagrams were taken as 0.5:9.5, 1:9, 2:8, 3:7, 4:6, 5:5, 6:4, 7:3, 8:2, 9:1, 9.5:0.5 for all the combinations. The required amount of the three components were weighed accurately and then sonicated for 3 minutes. The mixture was then

gently heated at 45–50ºC and vortex to form homogenous mixture. To this mixture distilled water was added drop by drop and mixture observed for 24 hour to check whether it remain transparent or become turbid. Minimum and maximum quantities of oil, surfactants and cosurfactants were optimized based on ternary phase diagrams.

Formulation development

Minimum 20 formulations F1 to F20 were prepared. Accurately weighed 20mg resveratrol was placed in glass vial and optimized combinations of oil, surfactant and co-surfactant were added. After adding all the components, the mixture was vortexd for few min and then homogenized at 45^{o}C for 5 mins. After homogenization, the mixture was sonicated in bath sonicator occasionally vortexes until drug was perfectly dissolved.

Evaluation

Percent drug content: The formulations F1 to F20 were filtered through membrane filter and then diluted with methanol. The concentration of resveratrol was then quantified using HPLC (Shimadzu, Kyoto, Japan) instrument equipped with two LC-10 ATVP pumps, SPD-10AVP UV-vis detector. Formulations having maximum percent drug content were further subjected to evaluation.

Determination of Zeta Potential: Zeta potential of optimized formulation was measured by photon correlation spectroscopy using Zetasizer (ZS nano, Malvern Instruments, United Kindom) with 4.0 mW He-Ne red laser (633nm) which measures potential range from -120 to 120V. Self-emulsion formulations (835mg) were diluted 100 times using distilled water and analyzed for zeta potential measurement at 25°C.

Transmission Electron Microscopic Analysis: The morphology of the optimized formulation was observed using TEM.[19,20] Optimized formulation was diluted with distilled water in ratio 1:200 and mixed by shaking. A drop of diluted formulation was applied to a 300-mesh copper grid and was left for 1 min. After this a drop of phosphotungstic acid (PTA) 2%w/v was applied to grid kept inverted for 10s. Excess of PTA was removed by absorbing on filter paper and grid was analyzed using JEM 2100F operated at 200 KV operated with AMT image capture instrument.

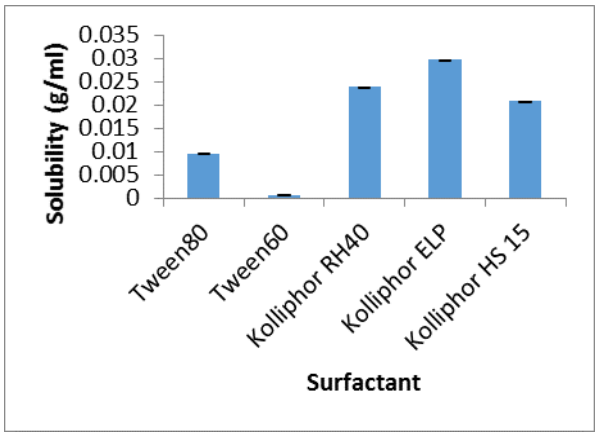
In vitro Resveratrol Release Study: Dissolution studies were carried out for optimized formulation, pure drug, marketed preparation in triplicate, employing USP Apparatus 2

(Labindia, Mumbai, India) with 900ml of 1.2 pH simulated gastric fluid containing 0.3% (w/v) sodium lauryl sulphate (SLS), stirred at 50rpm at a temperature of 37±0.5C [(1)]. At predetermined time intervals, an aliquot (1ml each) of the sample was collected, filtered, and analyzed for the content of RVT by the HPLC method.[14] An equivalent volume (1ml) of fresh dissolution medium was added to compensate for the loss due to sampling.

RESULTS

Solubility of Resveratrol in Lipids, Surfactant, Co-surfactants

Solubility studies were carried out to investigate the maximum soluble fraction of resveratrol in different lipids, surfactants and co-surfactants. The maximum solubility of resveratrol was observed in Labrafil M 2125 (0.04142 ± 0.000017g/ml), while the minimum was found in oleic acid (0.00002 ± 0.000015g/ml) (Figure 2**)** Higher solubility in lipids lead to lower requirement of surfactant and co surfactants, which reduce its toxic effects. Likewise, among the surfactants, the maximum solubility of RVT was observed in Kolliphor ELP (0.02972 ± 0.16g/ml) and minimum solubility was in Tween 60 (0.00063 ± 0.08g/ml) (Figure 3) and in cosurfactant maximum solubility was observed in Ethanol and PEG 400 (0.03383 ± 0.000222g/ml) and minimum solubility was found in Glycerin (0.00527 ± 0.000231g/ml) (Figure 4). Based on solubility studies oil Labrafil M 2125 and ethyl oleate was selected for further studies. Same, four surfactants (Kolliphor HS15, Kolliphor ELP, Kolliphor RH 40 and Tween80) and six co-surfactants (Ethanol, PEG 200, PEG400, n-butanol and Propylene Glycol) were selected respectively for further studies.



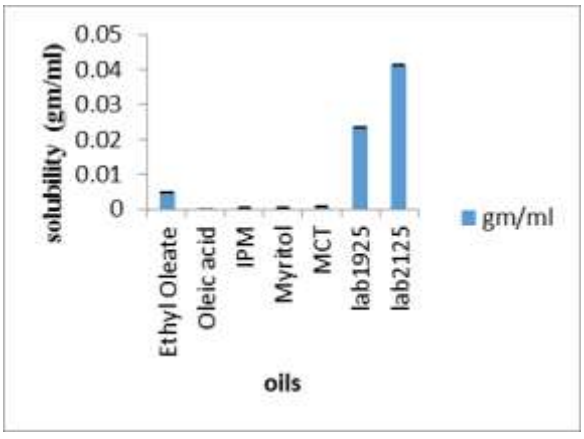


Fig 2: Solubility of drug in various oils. Fig 3: Solubility of drug in various surfactants.

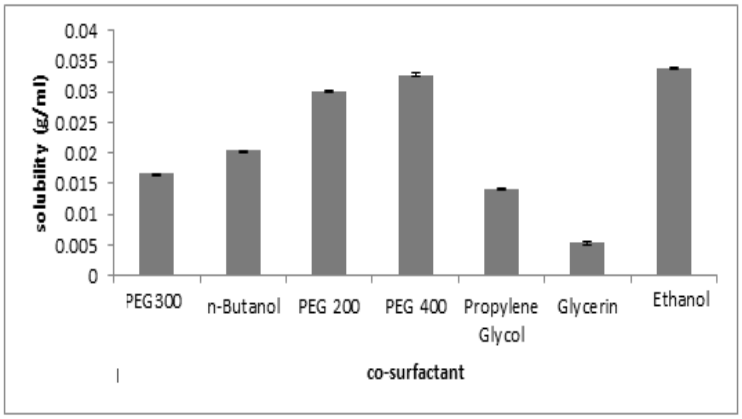


Fig 4: Solubility of drug in various co-surfactants.

Screening of Surfactants and Co surfactants

The selected oils, surfactants, cosurfactants were further subjected to screening, to find out best possible combination. During screening oil and surfactant were taken in ratio 1:1 as shown in table 2 formulation A1 to A8. Whereas during cosurfactant screening oil: surfactant: co-surfactant was taken in the ratios of 3:2:1 as shown in table 3 formulation B1 to B18. The ease of emulsification judged by the number of flask inversions required to yield homogeneous emulsion and % transmittance evaluated at 629 nm by using UV spectrophotometer as shown in Table 2,3. After clear observation of combination for 24hrs, three combinations i.e. combination 1: Labrafil M 2125+ Kolliphor HS15 + PEG 200, combination 2: Labrafil M 2125+ Kolliphor ELP + Ethanol and combination 3: Labrafil M 2125+ Kolliphor RH40 + PEG 400 were found to be the best and stable for formulation design of resveratrol as they remain transparent even after 24 hrs.

Table 2: Screening of Surfactants.

Formulation Code	Oils (mg)	Surfactants (mg)	Oil+Surfactant (mg) (1:1)	No. of Inversion	% Transparency
A1	Labrafil M 2125 (150mg)	Tween80 (150mg)	100mg	22 ± 1	25.2 ± 0.89
A2	Labrafil M 2125 (150mg)	Kolliphor RH 40 (150mg)	100mg	27± 2	98.2 ±0.54
A3	Labrafil M 2125 (150mg)	Kolliphor ELP (150mg)	100mg	18± 2	99.2 ±1.2
A4	Labrafil M 2125 (150mg)	Kolliphor HS15 (150mg)	100mg	32 ± 1	99.2 ±0.97
A5	Ethyl Oleate (150mg)	Kolliphor HS15 (150mg)	100mg	12 ± 3	49.2 ±0.65
A6	Ethyl Oleate (150mg)	Tween80 (150mg)	100mg	43 ± 4	52.2 ±0.74
A7	Ethyl Oleate (150mg)	Kolliphor RH 40 (150mg)	100mg	22 ± 2	93 ± 0.94
A8	Ethyl Oleate (150mg)	Kolliphor ELP (150mg)	100mg	16±2	94.6 ±0.99

Table 3: Screening of Co-Surfactants.

Form. code	Oil (mg)	Surfactant (mg)	Co-surfactant (mg)	Oil:surfactant: Cosurfactan (3:2:1)(mg)	No. of Inversion	% Transmittance
B1	Labrafil M 2125 (300)	Kolliphor RH 40 (200)	PEG 300 (100)	100	10± 2	81.4 ±0.89
B2	Labrafil M 2125 (300)	Kolliphor ELP (200)	PEG 300 (100)	100	4± 2	94.9 ± 0.54
B3	Labrafil M 2125 (300)	Kolliphor HS15 (200)	PEG300 (100)	100	13± 3	97.5 ± 0.64
B4	Labrafil M 2125 (300)	Kolliphor RH 40(200)	PEG200 (100)	100	3± 1	94.7 ± 0.94
B5	Labrafil M 2125 (300)	Kolliphor ELP (200)	PEG200 (100)	100	15± 3	95.8 ±0.79
B6	Labrafil M 2125 (300)	Kolliphor HS15 (200)	PEG200 (100)	100	3± 2	98.8 ±0.8
B7	Labrafil M 2125 (300)	Kolliphor ELP (200)	Ethanol (100)	100	5± 1	99.5 ± 0.87
B8	Labrafil M 2125 (300)	Kolliphor RH40 (200)	Ethanol (100)	100	80 ± 2	42.9 ±1.25
B9	Labrafil M 2125 (300)	Kolliphor HS 15 (200)	Ethanol (100)	100	7 ± 1	97.6 ±1
B10	Labrafil M 2125 (300)	Kolliphor RH 40 (200)	PEG 400 (100)	100	20 ± 2	99.5 ± 0.48
B11	Labrafil M 2125 (300)	Kolliphor ELP (200)	PEG 400 (100)	100	2 ± 3	83.8 ± 0.65
B12	Labrafil M 2125 (300)	Kolliphor HS 15 (200)	PEG 400 (100)	100	10 ± 2	95.9 ± 1.54
B13	Labrafil M 2125 (300)	Kolliphor RH 40 (200)	Propylene Glycol (100)	100	23 ±2	75.4 ± 0.78
B14	Labrafil M 2125 (300)	Kolliphor ELP (200)	Propylene Glycol (100)	100	5± 1	92 ± 0.69
B15	Labrafil M 2125 (300)	Kolliphor HS 15 (200)	Propylene Glycol (100)	100	3 ±1	95 ± 1.05
B16	Labrafil M 2125 (300)	Kolliphor RH 40 (200)	N-Butanol (100)	100	5 ± 1	95.3 ± 1.23
B17	Labrafil M 2125 (300)	Kolliphor ELP (200)	N-Butanol (100)	100	10 ±2	89.6 ± 0.87
B18	Labrafil M 2125 (300)	Kolliphor HS15 (200)	N-Butanol (100)	100	3 ± 1	61.8 ± 0.45

Optimization of oil, surfactant and cosurfactant combinations

The surfactant and co-surfactant were varied in mass ratios 1:1, 1:2, 2:1 for all the three combinations selected by screening. The different ratios of oil: Smix (surfactant and cosurfactant) as per pseudo ternary phase diagrams were taken as 0.5:9.5, 1:9, 2:8, 3:7, 4:6, 5:5, 6:4, 7:3, 8:2, 9:1, 9.5:0.5 for all the combinations as shown in figure 5. Out of three combinations, combination containing Labrafil 2125, Koliphor ELP and ethanol containing different ratio of surfactant and cosurfactant mixture showed maximum transparent region as observed from figure 5b. Thus, this combination was selected for formulation development. From ternary phase diagram calculated the percentage of oil, surfactant and cosurfactant, required to develop region of self-emulsion in which resveratrol particle size will remain in

nano size. Minimum and maximum percentage value of oil, surfactant, cosurfactant selected based on figure 5b, 5d are shown in table 4.

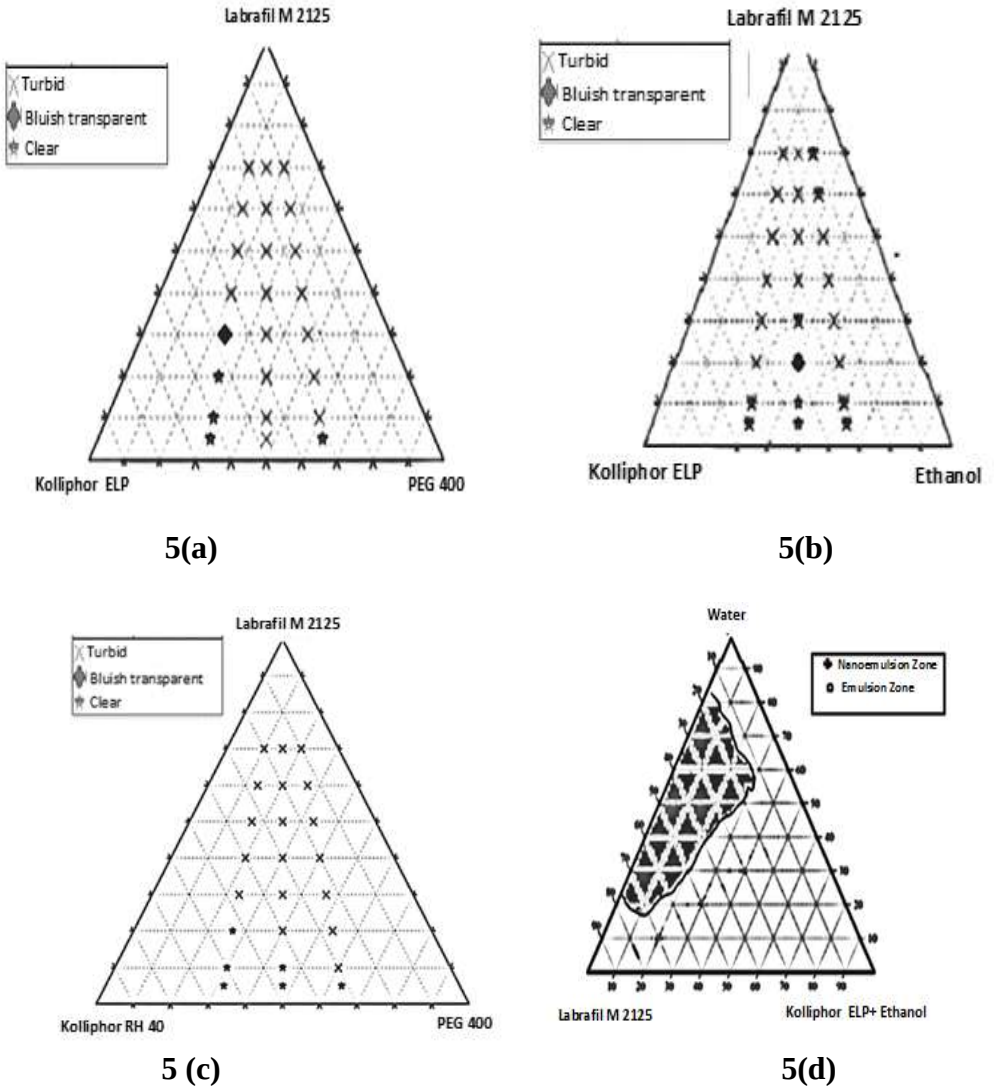


Fig. 5 Phase Diagrams Involving (a) Labrafil M 2125: Kolliphor HS15 (1:1) and PEG 200 as Cosurfactant, (b) Labrafil M 2125: Kolliphor ELP (1:2) and Ethanol as Cosurfactant, (c)Labrafil M 2125: Kolliphor RH40 (2:1) and PEG 400 as Cosurfactant. (d) Labrafil M 2125: Kolliphor ELP and Ethanol (S_{mix}): Water.

Formulation

Minimum 20 formulations F1 to F20 were prepared as shown in table 5 based on the minimum and maximum quantity selected from ternary phase diagrams (Table 4)

Table 4: Minimum and maximum quantity of ingridients.

Ingridient	Minimum (%w/w)	Maximum (%w/w)
Labrafil 2125	4.880	28.373
Kolliphor ELP	47.136	68.053
Ethanol	24.489	34.056

Evaluation

Percentage Drug content: The formulations were prepared by dissolving 20mg drug in optimized quantity of oil, surfactant and co surfactant from the ternary phase diagram as shown in table 5 by using vortex mixer and sonicator . Then the formulation was diluted with methanol and absorbance was measured at 306 nm by UV Spectrophotometer to calculate the percent drug dissolved. From table 5 it was found that F3, F14 had highest dissolved drug content.

Table 5: Percentage drug content of different formulation and transparency observation.

Formulation code	Oil % (Labrafil M 2125)	Surfactant % (Kolliphor ELP)	Co-surfactant % (Ethanol)	Percentage Drug content	Observation
F1	4.98	63.32	31.69	Drug not dissolved	Transparent and drug precipitated in less extent
F2	4.98	52.18	31.69	95.68±0.53	Transparent and drug precipitated in less extent
F3	4.98	52.18	26.01	99.71±0.52	Transparent and drug not precipitated
F4	21.81	52.18	31.69	80.48±0.88	Transparent and drug precipitated in less extent
F5	21.81	52.18	26.01	73.97±0.65	Transparent and drug precipitated in less extent
F6	13.395	67.12	28.85	81.578±0.29	Transparent and drug precipitated in less extent
F7	13.395	57.75	28.85	90.84±1.54	Transparent and drug

					precipitated in less extent
F8	13.395	57.75	24.07	Drug not dissolved	Turbid and drug precipitated in less extent
F9	13.395	57.75	28.85	89.57±0.58	Transparent and drug not precipitated
F10	4.98	63.32	26.01	88.51±0.49	Transparent and drug precipitated in less extent
F11	21.81	63.32	31.69	89.50±0.55	Transparent and drug precipitated in less extent
F12	13.395	57.75	28.85	91.94±0.61	Transparent and drug precipitated
F13	21.81	63.32	26.01	92.72 ±0.40	Transparent and drug precipitated
F14	13.395	57.75	33.62	100.59±0.58	Transparent and drug not precipitated
F15	-0.757	57.75	28.85	Not formed	Not formed
F16	13.395	57.75	28.85	89.40±0.85	Bluish transparent and drug precipitated in less extent
F17	13.395	48.382	28.85	90.89±0.56	Bluish transparent and drug precipitated
F18	27.54	57.75	28.85	89.51±0.69	Bluish turbid and drug precipitated
F19	13.395	57.75	28.85	82.75±0.19	Bluish turbid and drug precipitated
F20	13.395	57.75	28.85	68.27±0.94	Bluish turbid and drug precipitated

Transparency and Drug Precipitation: The prepared formulation was diluted up to 5 ml and observed visually for the transparency and drug precipitation. It was observed from table 5 that F3 and F14 remain transparent even after 24 hrs. These two formulations were subjected to further evaluation.

PH of Formulations: pH of both formulations was found to be in a range of 6.3±0.64 to 6.4±0.29.

Determination of the emulsification time: To determine the emulsification time, formulation was added to 200 ml & 900ml of 0.1N HCl. at 37°C with gentle agitation using a

magnetic stirrer. The formulation was assessed visually per the rate of emulsification and the final appearance of the emulsion observed as shown in table 6.

Table 6: Emulsification time and appearance of Self Emulsion Formulation.

S. No.	Formulation Code	Emulsification time	Appearance in 200ml		Appearance in 900ml	
			After dilution	After 24 Hr.	After dilution	After 24 Hr.
1	F3	Within 3sec.	Homogenous, Clear transparent	Homogenous, Clear transparent	Homogenous, Clear transparent	Homogenous, Clear transparent
2	F14	Within 5-6sec.	Homogenous, Bluish Transparent	Turbid	Homogenous, Bluish Transparent	Turbid

Determination of globule size and zeta potential: The zeta potential of formulation F_3 was found to be 21.3mV zeta potential (Figure 6a) and globule size was found to be 83.25nm (Figure 6c) and zeta potential of formulations F_{14} was found to be -19.5mV zeta potential (Figure 6b) and globule size was found to be 172.2nm (Figure 6d)**.** From these observations, it was found that F3 formulation has smaller size and greater potential. So, it was further subjected to TEM analysis.

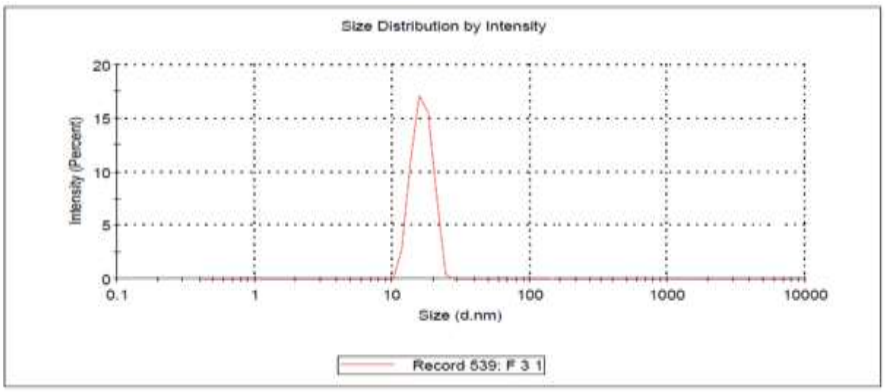


Fig 6a: Graph of Particle Size of F3 Formulation.

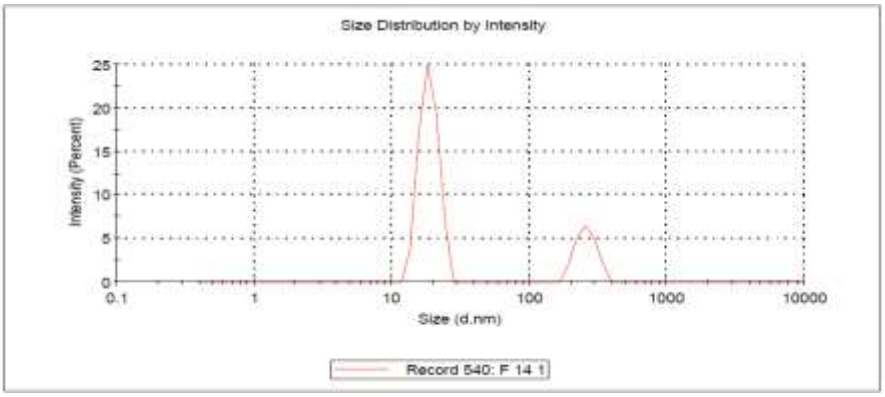


Fig 6b: Graph of Particle Size of F14 Formulation.

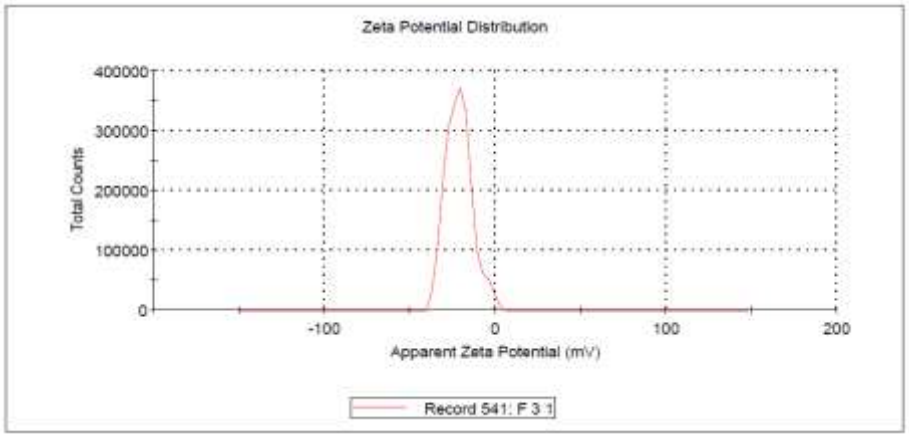


Fig 6c: Graph of Zeta Potential of F3 Formulation.

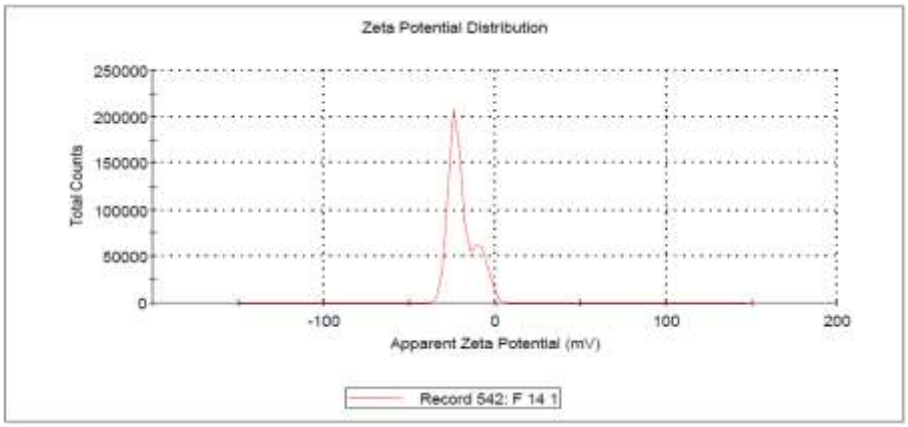


Fig 6d: Graph of Zeta Potential of F14 Formulation.

Transmission Electron Microscopic Analysis: Transmission electron microscopic (TEM) images of formulations F_3 after 24 h of post dilution in distilled water (Figure 7**).** It was observed that spherical micelles had no signs of coalescence even after 24h of post dilution. The nanoemulsion droplets emerged as dark and the surroundings were observed inferring the stability of formed nanoemulsion. Closer images reveal that each globule is surrounded by thick layer. This indicates formation of monolayer around the emulsion droplets, reducing the interfacial energy. Image shows the electron microscopic image, depicting the morphology of the reconstituted optimized formulation., all the globules were of uniform shape, with globule size of most of them was less than 100nm. The figure clearly illustrates that there are no indications of coalescence, so physical stability of the formulation was enhanced.

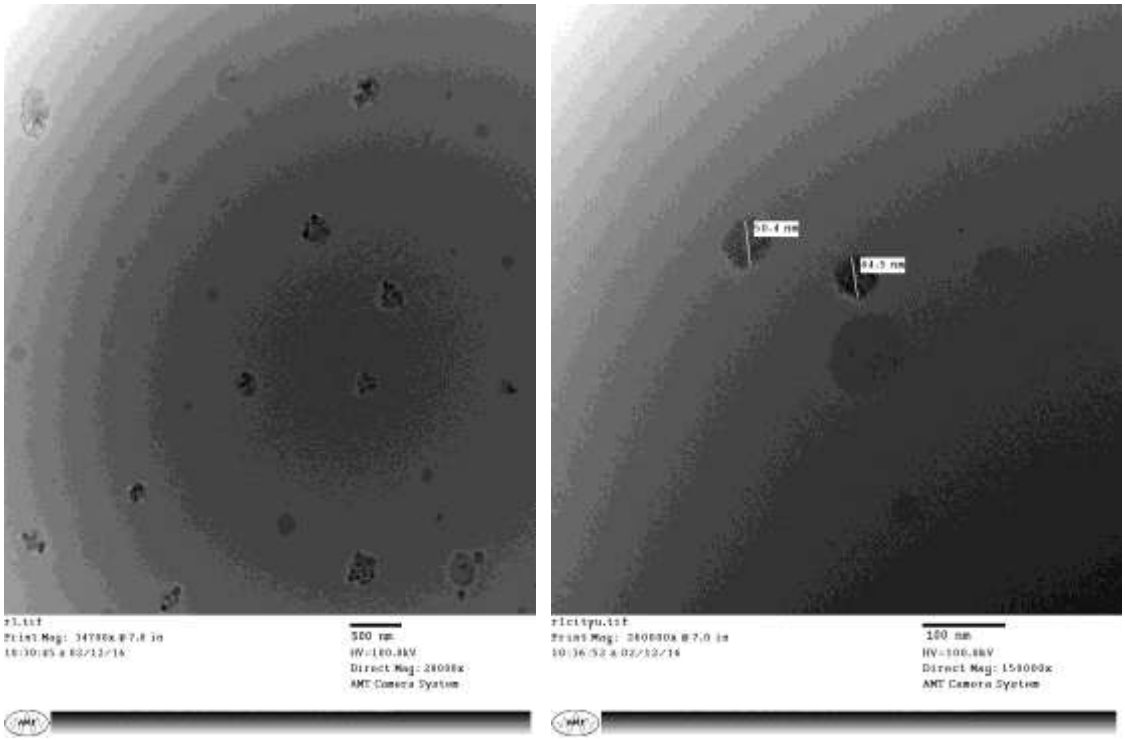


Fig 7: TEM Images of Resveratrol Loaded SNEDDS Formulation F3.

***In vitro* Resveratrol Release Study:** Resveratrol release from formulation F_3 was compared with pure resveratrol and marketed resveratrol powder formulation. Release was observed to be significantly improved in self- emulsion formulation **(**Figure 8**).** Drug dissolution was nearly completed within 30min in case of the optimized formulation, as compared with that of the pure drug and marketed preparation where drug release was less than 50% in 30mins.

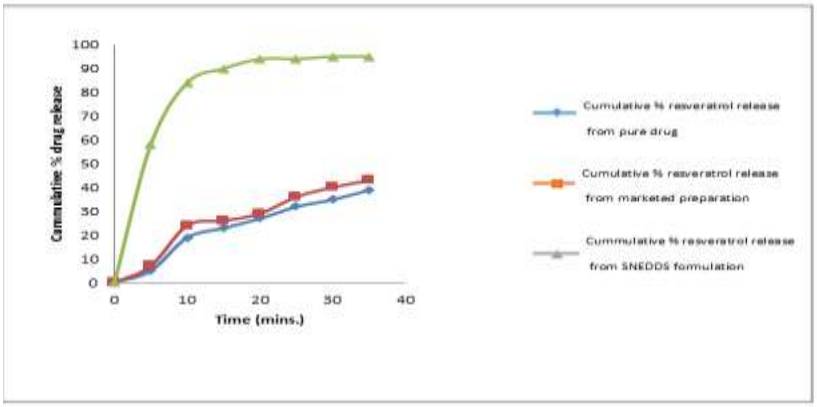


Fig 8: *In vitro* Resveratrol Release Study.

DISCUSSION

Solubility studies were performed to find out the maximum solubility of resveratrol in different lipids, surfactants and co-surfactants. Results obtained clearly indicate that resveratrol was having maximum solubility in Labrafil M 2125 which was selected as the

lipid excipient for formulation development. Labrafil M 2125 is a known as bioavailability improving agent which enhances oral bioavailability by inhibiting enzyme CYP3A4 which is an enterocyte drug-metabolizing enzyme. Non-ionic surfactants are known to have less toxicity than ionic surfactants. Therefore, usually accepted for oral formulations. In addition, they are known to produce reversible changes in mucosa of intestine, thus lead to change permeability of lipophilic drugs. Therefore, Kolliphor ELP was selected as surfactant in which resveratrol showed maximum solubility, whereas ethanol was selected as cosurfactant in the formulation development with an objective to enhance drug-loading efficiency. Literature clearly reports that HLB value of surfactant lower interfacial energy, which leads to formation of a stable emulsion. The surfactant added in formulation, i.e. Kolliphor ELP, has HLB value of 15. Thus, HLB values and ternary plots obtained indicated the synergistic effect in reducing interfacial tension and formation of stable nanoemulsion. The nanoemulsion droplets emerged as dark and the surroundings were observed inferring the stability of formed nanoemulsion. Closer images in TEM reveal that each globule is surrounded by thick layer. This indicates formation of monolayer around the emulsion droplets with reduced interfacial energy. Significantly high dissolution rate of self- emulsion formulation could be attributed to small globule size of 83.25 nm, high percentage transmittance (99.5 ± 0.87), negative zeta potential and in situ solublization as confirmed by TEM studies, which provided large surface area for release of drug and thus permitting faster rate of drug release.

CONCLUSION

The study demonstrated that HLB value, oil solubility, hydrophobic chain length, greatly influence microemulsion and nanoemulsion. The study demonstrated that solid lipid nanoparticle, self-emulsifying drug delivery system, liposome formulation will be the promising strategies to enhance solubility, stability and in vitro release of resveratrol. The spontaneous development of nanoemulsion presents the drug in dissolved form. The nano size droplet formed provides large interfacial surface area for drug absorption. In addition, the specific nanoemulsion will promote intestinal transport of drug through lymphatic transport of system.

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DISCLOSURE

The report has no conflicts of interest in this work.

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