FORMULATION OPTIMIZATION AND CHARACTERIZATION OF SELF-MICRO EMULSIFYING DRUG DELIVERY SYSTEM OF IVERMECTIN

SYNOPSIS OF THE THESIS

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In partial fulfilment of the requirements for the award of the degree of

MASTER OF PHARMACY

IN

PHARMACEUTICS

Submitted by

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DEPARTMENT OF PHARMACEUTICS
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CERTIFICATE

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Place: Chennai-03

Date:

(Dr. A. JERAD SURESH, M. Pharm., Ph.D., M.B.A)

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Place: Chennai-03

Date:

(Mr. K..Ramesh kumar, M. Pharm..)

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“Gratitude makes sense of our past, brings peace for today and creates a vision for tomorrow”

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TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

ABBREVIATIONS USED

HTS	High Throughput Screening
BCS	Biological Classification System
PWSDs	Poorly Water Soluble Drugs
LBDDS	Lipid Based Drug Delivery System
SLN	Solid Lipid Nanoparticles
NLC	Nano Structured Lipid Carriers
CS	Cholesterol
TG	Triglyccerides
DG	Diglycerides
FA	Fatty acids
PL	Phospholipids
LC	Liquid Crystals
BS	Bile Salts
HGL	Human Gastric Lipase
HPL	Human Pancreatic Lipase
CEH	Carboxyl Ester Hydrolase
DE	Diesters
ME	Monoesters
DGDG	Digalactosyldiglycerides
DGMG	Digalactosylmonoglycerides
UWL	Unstirred Water Layer
VLDL	Very Low Density Lipid
LFCS	Lipid Formulation Classification System
GIT	Gastro Intestinal Tract
HLB	Hydrophilic Lipophilic Balance
O/W	Oil in water
W/O	Water in oil
SEDDS	Self Emulsifying Drug Delivery System
SMEDDS	Self Micro Emulsifying Drug Delivery System
SDEDDS	Self Double Emulsifying Drug Delivery System
SNEDDS	Self Nano Emulsifying Drug Delivery System
S-SNEDDS	Solid Self Nano Emulsifying Drug Delivery System
BHT	Butylated Hydroxy Toluene
BHA	Butylated Hydroxy Anisole
PG	Propyl Gallate
CAT	Compartment Absorption Model
P.eff	Permeability Co-efficient
PDS	Plain Drug Suspension
MF	Marketed Formulation

PDI	Poly Dispersibility Index
%T	% Transmittance
MDR	Mean Dissolution Rate
C Max	Concentration Maximum
SEM	Scanning Electron Microscope
XRD	X- Ray Diffraction
DSC	Differential Scanning Calorimetry
API	Active Pharmaceutical Ingredient
CHF	Congestive Heart Failure
FTIR	Fourier Transform Infra Red
BP	British Pharmacopoeia
IP	Indian Pharmacopoeia
JP	Japan Pharmacopoeia
PhEur	European Pharmacopoeia
USP NF	United States Pharmacopoeia, National Formulary
RI	Refractive Index
UV	Ultra violet
RPM	Revolution Per Minute
Df	Degree of Freedom
P-Value	Probability value
F	Distribution under the null hypothesis
CI	Confidence Interval
SE.Mean	Standard Error Mean
VIF	Variance Inflation Factor
PRESS	Predicted Residual Error Sum of Squares
SD	Standard Deviation
AST	Accelerated Stability Testing
ICH	International Council on Harmonization
EDTA	Ethylene Diamine Tetra Acetate
MCT	Medium Chain Triglycerides
LCT	Long Chain Triglycerides
MCM	Medium Chain Monoglycerides
P-gp	P-glycoprotein
GRAS	Generally Regarded as Safe
CMC	Critical Miceller Concentration
CEH	Carboxyl Ester Hydrolase
FaSSIF	Fasted State Stimulated Intestinal Fluid
FeSSIF	Fed State Stimulated Intestinal Fluid
NDA	New Drug Application
MAA	Marketing Authorization Application

SYMBOLS USED

cps	Centipoise
nm	Nanometre
cm	Centimetre
mV	Milli volt
kV	Kilo Volt
mA	Milliampere
μg	Microgram
ml	Millilitre
λ_{max}	Maximum Wavelength of Absorbtion
mg	Milligram
g	Gram
sec	Second
min	Minute
hrs	Hours
ζ	Zeta Potential
%	Percentage
^0C	Degree Celsius
t	Time
k^0	Zero Order Constant
C	Concentration
h	Height
r	Radius

INTRODUCTION

1. INTRODUCTION

In the last 2 decades, many tools in drug discovery and screening have been rapidly developed. Examples are automated synthesis, combinatorial chemistry, molecular genetics and high- throughput screening (HTS) methodologies. Accordingly, a large number of compounds have been identified as potential drug candidates[1-4]. Lipinski et al.[1, 2] have proposed the "rule of 5" to identify the potential poorly bio available drug candidates. Proposed properties of poor bioavailability include: (a) high molecular weight (> 500 D), (b) high lipophilicity (Log P > 5 or MLogP > 4.15), (c) possession of more than 5 H-bond donors (e.g. NHs and OHs) and (d) possession of more than 10 H-bond acceptors (e.g. Ns and Os). This rule is only valid for drug candidates that are not substrates for active transporters and efflux mechanisms. Poor bioavailability always originates from poor aqueous solubility or poor intestinal permeability[5].

Amidon et al.[6] have introduced the Biopharmaceutical classification system (BCS). BCS classifies drugs according to their maximum dose solubility, dissolution and permeability into four classes (Fig 1). High solubility means that the maximum dose is soluble in 250 ml aqueous media in the pH range of 1-7.5 at 37°C. High dissolution means that not less than 85 % of the administered dose is released within 30 min. High permeability means that more than 90 % of the dose is absorbed. This classification provides a guiding tool to replace individual bioequivalence studies by accurate in vitro dissolution tests[7, 8].

Unfortunately, the number of potential drug candidates, especially those with high molecular weight and high Log P, is progressively increasing. Accordingly, the problem of the poor aqueous solubility (< 1 µg/ml) has become dominant in the pharmaceutical industry[1, 2, 9] .Recent studies showed that ~75 % of the drug development candidates are poorly water-soluble. This ratio could be increased to 80-90 % depending on the therapeutic area [8, 9]. Poorly water-soluble drugs (PWSDs) represent Class II and IV of the BCS6.

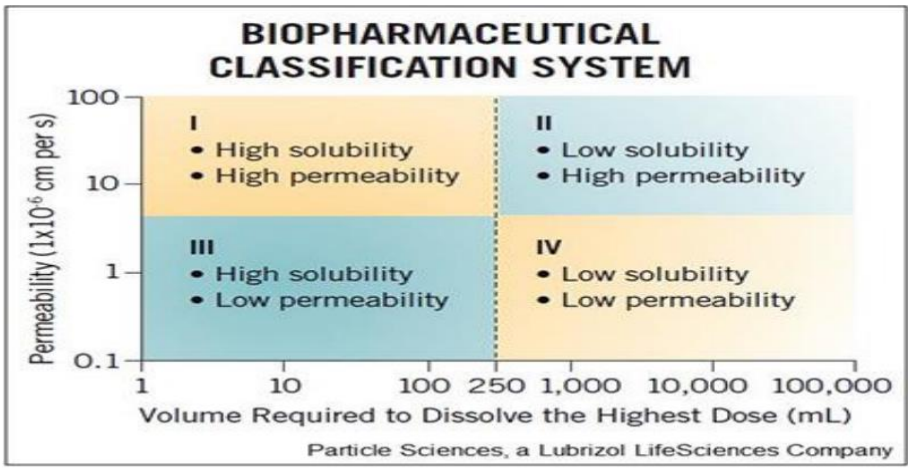


Fig 1. Biopharmaceutical classification system of drugs (BCS).

Several strategies have been developed to enhance the water solubility and hereafter the bioavailability of PWSDs. These strategies could be briefly summarized into: (a) Physical modifications such as particle size reduction, optimization of crystal habit, co-crystal formation and solid dispersions. (b) Chemical modifications such as the use of buffers, salt formation and complexation (Cyclodextrins). (c) Miscellaneous methods such as the use of surfactants, co-solvents, hydrotrophy, supercritical fluids and lipid-based drug delivery systems (LBDDS) [3, 4, 7,13] .

Lipids represent a large class of compounds that can be classified according to their chemical structures, origin and solubility in organic solvents or biochemical interactions[14]. A pioneer in the field of lipid-based systems, Small [15] has introduced a lipid classification system based on lipid/water interactions in bulk water and the behaviour of lipids at the air/water interface (Table 1).

Table 1. Lipid classification system proposed by Small [15].

Class	Bulk interactions with water	Surface interactions with water	Examples
Non-polar	- Insoluble - Crystals or oil	Do not spread to form a monolayer	Cholestanes, benzpyrenes, carotenes, lycopenes and gadusenes
Polar I	- Insoluble, non-swelling - Crystals or oil	Form a stable monolayer	CS, TG, DG, long chain protonated FA, waxes, sterols, oil soluble vitamins and steroidal hormones
Polar II	- Insoluble, swelling - LC	Form a stable monolayer	PL, MG, FA soaps and cerebrosides
Polar IIIA	- Soluble with lyotropic mesomorphism -Crystals or oil micelles	Form an unstable monolayer	Lysolecithins and surfactants
Polar IIIB	- Soluble without lyotropic mesomorphism micelles - Crystals or oil	Form an unstable monolayer	BS and saponins

CS: cholesterols; TG: triglycerides; DG: diglycerides; FA: fatty acids; PL: phospholipids; LC: liquid crystals; BS: bile salts.

LBDDS present and maintain the drug in the solubilised form, in which absorption takes place[22, 23]. As a result, the rate-limiting step of drug dissolution is eliminated. Furthermore, they can enhance the bioavailability by different mechanisms depending on their type and amounts such as prolongation of the gastric emptying time; stimulation of bile secretion and interaction with bile salts (BS), phospholipids (PL) and cholesterol (CS) mixed micelles; reduction of the first pass metabolism via stimulation of intestinal lymphatic transport for highly lipophilic drugs (Log P > 5) and reduction of the enterocyte-based metabolism; modulation of intestinal efflux transporters such as P-glycoprotein; permeation enhancement

as well as generation and maintenance of a metastable supersaturable drug state[23-25] . Oral administration of lipids stimulates the secretion of the gastric lipase (HGL) with the consequent secretion of the pancreatic lipase (HPL) and co-lipase from the pancreas along with other esterase's such as phospholipase A2 (PLA2), carboxyl ester hydrolase (CEH) and pancreatic lipase related protein 2 (PLRP2)[26,27] . Most of the lipid excipients are esters. Examples are glycerides, PEG esters of fatty acids, polysorbates, PL and CS esters. Ester bonds are generally potential substrates to lipolytic enzymes.

Lipid digestion usually starts in the stomach by the action of HGL[28]. HGL is an acid stable lipase with an optimum activity at pH 3-6 and a maximum activity at pH 5.0-5.425, [27]. HGL is secreted by the chief cells of the gastric fundic glands under the stimulation of meals, stomach motion, gastrin secretion and cholinergic mechanisms[26, 29]. HGL works on the lipid/water interface. Therefore, the ingested lipids need to be emulsified before being digested. The emulsification is usually achieved by the shear action of the stomach along with the surface active actions of the co-administered amphiphiles and digestion products such as monoglycerides (MG) and dietary protein[27].

Therefore, the contribution of the gastric lipolysis to the whole lipid digestion process is strongly dependent on the gastric residence time, susceptibility of the ingested lipid to digestion and lipid dispersibility pattern in the gastric fluids[23]. In some cases such as incomplete pancreatic function (neonates) or compromised one (cystic fibrosis or chronic alcoholism), gastric lipolysis plays the principal role in the lipid digestion[28, 30]. However, in most cases, gastric lipolysis accounts only for 10-25 % of the total lipid lipolysis[31, 32]. For example, triglycerides (TG) could be partially hydrolyzed in the stomach into diglycerides (DG) and free fatty acids (FA) [28, 31, 33]. FA is protonated under the gastric conditions. In the absence of the bile mixed micelles, protonated FA (especially long chain ones) accumulate on the lipid/water interface with subsequent deactivation of the HGL[27, 29, 30]. HPL is produced in the acinar cells of the pancreas and is secreted along with bile under the stimulation of cholecystokinin and secretin.

HPL is active only above pH 5 with a maximum activity at pH 7.0-7.5[29]. Similar to HGL, HPL works on the lipid/water interface. However, BS always desorbs HPL from the interface with the subsequent inhibition of its action[34]. This inhibitory effect is counter balanced by the formation of HPL/co-lipase equimolar complex, which plays a crucial role in the HPL anchoring to the lipid/water interface. Furthermore, FA produced during the lipolysis enhances the anchoring of HPL/co-lipase complex on the lipid/water interface with the consequent promotion of further lipid lipolysis[35]. In addition, FA indirectly stimulates the HPL secretion through cholecystokinin release stimulation[36, 37]. Moreover, the presence of the lipid digestion products, especially long chain FA in the small intestine is reported to reduce the gastric motility with the subsequent delay of the gastric emptying rate[38].

This delay may allow more efficient lipid lipolysis and absorption in the upper GI tract as well as higher PWSD dissolution. Both HGL and HPL have high selectivity toward TG. However, they differ in their specificity. HPL is a region selective enzyme that hydrolyzes only sn-1 or sn-3 positions. On the other hand, HGL can hydrolyze the 3 ester positions[29].

Other lipolytic enzymes such as CEH, PLA2 and PLRP2 do not work on the interface. They hydrolyze the lipid excipients in the dispersed micelles or mixed micelles[27]. Therefore, they are beneficial in the digestion of various lipid excipients[39].

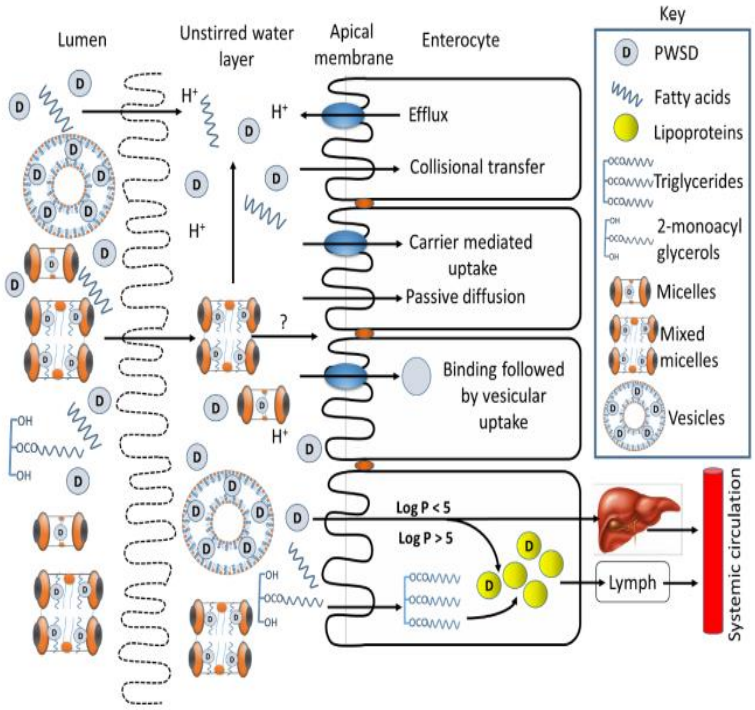


Fig 2. Schematic representation of different mechanism of lipids and drugs absorption as well as lipid-mediated bioavailability enhancement[27].

Self-emulsifying drug delivery systems represent class II and class III of the LFCS (Lipid formulation classification system (LFCS) proposed by Pouton). They are composed of two or more ingredients, which provide the self-emulsifying properties: more hydrophilic amphiphiles, more lipophilic amphiphiles and sometimes co-solvents or precipitation inhibitors. Upon mild agitation and dilution in the GI fluids, these systems transform into oil in water (O/W) emulsions (SEDDS), double emulsions (SDEDDS), micro emulsions (SMEDDS) or nanoemulsions (SNEDDS) [13, 43]. Micro emulsions are thermodynamically stable while nanoemulsions are only kinetically stable. However, in most of the literatures, SNEDDS and SMEDDS are usually subjectively assigned to formulations that provide fine colloidal dispersions.

Self-emulsification increases the bioavailability by the circumvention of drug crystal dissolution, which is often insufficient and highly variable for the PWSDs[45]. Compared to the conventional emulsions, SNEDDS are water-free systems. Accordingly, they have better physical and chemical stability. SNEDDS have high patient compliance and palatability as they are always formulated as capsules or tablets. Food has minor effect on drug absorption from SNEDDS compared to other LBDDS. Other advantages include the ease of manufacture and scale-up as well as quick onset of action[16]. In addition, being a mixture of more lipophilic

and more hydrophilic amphiphiles, SNEDDS offer high solubilisation capacity to a wide spectrum of PWSDs with different degrees of lipophilicity compared to other LBDDS[46].

A good example of a very successful formulation is Neoral® / Optoral® (Novartis). It is composed of a mixture of MG, DG and TG as lipophilic amphiphiles, Cremophore® RH 40 as a hydrophilic amphiphile, propylene glycol and ethanol as co-solvents and tocopherol as an antioxidant[42]. It forms spontaneously transparent dispersions with particles size below 100 nm upon dilution with aqueous media[47]. SNEDDS are not only restricted for the oral use[48-51]. Self-emulsifying suppositories[52], intraurethral liquid formulations[53], injections[54], implants[55], transdermal[56] and ocular systems[57] are also reported. The mechanism of the self-emulsification process is still not clear. However, Reiss[58] has suggested that self-emulsification occurs when the entropy change in the favour of dispersion is higher than the energy required to increase the surface area of the dispersion. The free energy of an emulsion is a function of the energy required to create a new surface between the oil and water phases that could be described by the following equation:

$$\Delta G = \sum N\pi r2\ \sigma$$

where ΔG is the free energy associated with the process, N is the number of droplets, r is the radius of the droplets and σ is the interfacial energy. The free energy of mixing is ignored.

Crude emulsions are not thermodynamically stable. Therefore, oil and water phases have a high tendency to separate in order to reduce the interfacial energy. The presence of the more hydrophilic amphiphiles stabilizes the interface and reduces the interfacial free energy by formation of a monolayer around the oil droplets. In the case of the SNEDDS, the free energy required to form the emulsion is very small and could be positive or negative. Therefore, the emulsification process takes place spontaneously[49]. The easiness of the emulsification was proposed to be related to the ease of water penetration into the various LC or gel phases formed on the surface of the droplets[18]. The interface between the oil and the aqueous continuous phase is formed upon addition of the oil/hydrophilic amphiphiles mixture to the water. Water penetrates then into the interface and is solubilised in the oil phase. The extent of water penetration is dependent on its solubilisation limit close to the interface[58]. Further aqueous penetration leads to the dispersion of the LC phase. Finally, oil droplets surrounded by LC interface are formed. The extent of the LC interface depends on the hydrophilic amphiphile concentration in the mixture21[46].

Several lipid excipients could be formulated as SNEDDS[20, 24, 59]. Based on their polarity, HLB and interaction with the aqueous media, they could be classified as more lipophilic amphiphiles (Polar lipids I and II) and more hydrophilic amphiphiles (Polar lipids IIIa). There are several factors that should be considered in the selection of the lipid excipients. The most important factor is toxicity, especially if the SNEDDS are intended for chronic use. Other factors include the solvent capacity, melting point, digestibility, capsule compatibility, chemical stability, purity, miscibility and their role in promoting the self-dispersibility[17]. More hydrophilic amphiphiles lead to formations, which readily disperse. However, they show in many cases low drug loads and are sensitive to dilution. If the content of the more

lipophilic amphiphiles is increased, often higher drug loads can be achieved. However, the self- nanoemulsifying properties are decreased. Therefore, a balanced composition is crucial for the in vivo performance.

Advantages of SMEDDS over other emulsions[1-9]:

1. Storage: SMEDDS has the same advantage as emulsions, of facilitating the solubility of hydrophobic drugs. Macro emulsions undergo creaming over a period of time, whereas SMEDDS being thermodynamically stable can be stored easily.

2. Stability: In contrast to micro/nanoemulsions, SMEDDS do not contain water and hence, they have improved physical and/or chemical stability on long-term storage. Self-nanoemulsifying tablets of carvedilol showed successful incorporation of carvedilol within the SNEDDS. This resulted in improvement of the stability of carvedilol on dilution with aqueous media in the presence of cellulosic polymers.

3. Compliance: Most of the SMEDDS formulations are in capsule or tablet dosage forms, thus occupying smaller volume, easy to administer and hence improved patient compliance.

4. Palatability: SMEDDS formulation can be easily filled into capsules resolving the palatability issues associated with lipid formulations.

5. Effect of food: Absorption of drug from SMEDDS formulation is not affected by food. The lipophilic contents of fatty diet aid, aids in absorption of drug from these systems. It was observed that food had a marked effect on the absorption of itraconazole from the marketed formulation (Sporanox capsule), whereas the influence was less pronounced for the self-emulsifying formulation of itraconazole (ITRA-GSMP capsule) in human volunteers.

6. Quick onset of action: SMEDDS have the ability to facilitate rapid oral absorption of the drug, which results in quick onset of action. It was found that the t_{max} al., of vitamin A was reduced and bioavailability was increased when administered as SNEDDS capsule and SNEDDS tablet as compared to vitamin A oily solution-filled capsules without any additives.

7. Ease of manufacture and scale-up: SMEDDS can be easily manufactured at large scale as it requires simple and economical manufacturing facilities, such as simple mixer with an agitator and volumetric liquid filling equipment.

Limitations of SMEDDS

Although SMEDDS formulation has several advantages, there are certain limitations associated with this system

SNEDDS are associated with some limitations[45] that need to be considered during the formulation development and manufacture. Examples are:

(a) The susceptibility of some lipids to oxidation and polymorphism. Lipid oxidation could be reduced by the use of saturated lipids or the incorporation of antioxidants or metal chelators such as EDTA[64]. Polymorphism is always associated with long chain lipids. The influence of the polymorphism can be avoided by heating the lipids at least 20 °C above their melting point and good homogenization. This approach destroys any preformed crystals and promotes the uniformity of the solidified product.

(b) PWSDs precipitation upon dilution. SNEDDS, especially those with high co-solvents content, carry high risk of PWSD precipitation upon dilution due to the loss of solubilisation capacity[7]. The degree of precipitation depends on the lipophilicity of the PWSDs as well as the contribution of the hydrophilic amphiphiles and co-solvents to the PWSDs solubilisation. However, the precipitation kinetics could be in some cases very slow so that the PWSDs remain in the supersaturated state for a considerable time. Accordingly, the in vivo absorption of the PWSDs is not pronouncedly affected[16]. Furthermore, precipitation could be reduced by incorporation of precipitation inhibitors such as HPMC to provide and maintain metastable supersaturated drug state.

(c) SMEDDS can only accommodate low drug dosage. However, the SNEDDS-mediated PWSD bioavailability enhancement may outweigh the dose reduction[16].

(d) SMEDDS are typically formulated as liquid to be encapsulated in soft gelatin capsules. Several drawbacks are associated with such systems such as the interaction with the capsule shell, capsule leakage, instability, possible drug precipitation upon temperature variation as well as the requirement of specialized manufacturing equipment[66].

Therefore, alternative formulation strategies, e.g. the inclusion of the SNEDDS into a solid (S- SNEDDS) or semisolid dosage form, are desirable, nonetheless, very challenging. S-SNEDDS combines the benefits of liquid SNEDDS with those of solid dosage forms and overcomes its limitations. S-SNEDDS were formulated as

- pellets[67,68],
- conventional tablets[69] ,
- bilayer tablets[70],
- effervescent tablets[71],
- orodispersible tablets[72],
- capsules[73],
- tablet-loaded pulsatile capsules[74],
- osmotic pumps[49,60],
- microparticles[75],
- nanoparticles[76],
- mouth dissolving films[77],
- beads[78],
- lipid matrices and self-emulsifying glasses[61, 62].

Several approaches were evaluated for the manufacture of the S-SNEDDS. These approaches could be summarized into: [80-82]

1. The use of solid or semisolid lipids.
   Liquid, semisolid and/or solid lipids could be blended so that the final form would have a semisolid or solid consistency. Examples of the evaluated semisolid/solid lipids are: Acconon® C-44, Acconon® C-50, Gelucire® 50/13 and Gelucire® 44/14. Compared to other approaches, higher lipid/drug load and scale up simplicity is afforded. Nevertheless, this approach is very challenging because self-nanoemulsifying properties are harder to be achieved with solid lipids. Furthermore, PWSDs could be crystallized out when the molten solid lipids reach room temperature.
2. Incorporation of polymeric excipients/amphiphiles.
   The liquid SNEDDS is homogeneously distributed in a hydrophilic polymeric matrix such as PEG. Alternatively, solid polymeric amphiphiles such as Poloxamer could be used to prepare S-SNEDDS. Poloxamer 188 plays a dual role, as a solidifying agent and more hydrophilic amphiphiles, in the production of S-SNEDDS. In both approaches (1 and 2), the solidified lipids could be directly filled into capsules in the molten state or transformed into powders using cryogenic grinding, melt granulation or spray cooling (congealing). The produced powders could be filled into hard gelatin capsules or compressed into tablets.
3. Lyophilisation.
   The aqueous phase is removed from O/W emulsions by freeze drying to produce dry emulsions. Direct lyophilisation in suitable PVC blisters could be used to prepare self-nanoemulsifying tablets.
4. Extrusion/sphereonization.
   The liquid SNEDDS is mixed with a pelletization aid such as MCC and lactose. The produced mass is extruded and spheronized into freely flowable pellets.
5. Adsorption onto solid carrier.
   Liquid SNEDDS are adsorbed onto porous carriers e.g. silicates to prepare apparently dry freely flowable powders. Ideal adsorbent should not interfere with the self-nanoemulsifying properties, have higher adsorption capacity, have superior flow properties, able to produce tablets with acceptable physical properties and able to release 100 % of the incorporated SNEDDS/PWSDs. Several silicates with different physical properties and pore sizes were evaluated. Examples are: Aeroperl® 300, Aerosil® 200, Neosyl®, Neusilin® UFL2, Neusilin® US2, Sipernat®, Sylysia® and Zeopharm®. Other evaluated non-silicates adsorbents include Fujicalin®, Hydroxypropyl-β-cyclodextrin, Magnesium stearate, Mannitol, MCC, Polyvinyl alcohol and Sodium carboxymethyl cellulose.
   The adsorption process could be performed by:
   (a) Solvent-free methods. The adsorption process could be done by simple triturating in mortar using a pestle or using a mechanical mixer. Alternatively, the liquid SNEDDS/adsorbent mixture could be wet granulated to produce freely flowable granules

(b) SNEDDS are dissolved in an organic solvent or emulsified in water. The SNEDDS solutions/emulsions are then mixed with the adsorbent and the aqueous/organic phase is removed by rotary evaporation, spray drying or freeze drying

6. Liquisolid technique.
   The carrier, usually MCC, is saturated with the liquid SNEDDS. Excess surface liquid is coated with silicates to produce apparently dry, freely flowable powders
7. Fluid bed coating.
   Porous silicates/MCC pellets are prepared by extrusion/spheronization. The liquid SNEDDS is then sprayed onto the surface of the porous pellets in a fluid bed coater. Alternatively, liquid SNEDDS are emulsified in water and mixed with a film former such as Poly vinyl pyrrolidone K30. Non-pareil pellets are then coated with the mixture in a fluid bed coater. In all cases, the prepared S-SNEDDS powder could be filled into capsules118 or compressed into tablets. Although S-SNEDDS are less reactive with the capsule shell than the liquid ones, shell softening is still observed in some cases upon storage. HPMC capsules are superior upon storage of S-SNEDDS compared to hard gelatin ones. Furthermore, due to the relatively low density of SNEDDS adsorbents, tablets are more favourable than capsules. Tablets can hold 2-3 times more powder compared to capsules.
   However, compression of SNEDDS-loaded adsorbents is not trivial. The SNEDDS could be squeezed out during the compression. Furthermore, the hydrophobic environment inside the produced tablets hinder their disintegration and can lead to incomplete drug release, especially when gel-mediated SNEDDS dispersion takes place or irreversible interaction between the SNEDDS and adsorbent arises.

   Several approaches were explored to prepare S-SNEDDS tablets. Examples are lyophilisation, wet granulation, dry granulation and direct compression. Alternative approach is to prepare plain tablets with high porosity. Subsequently, tablets are loaded by soaking into the liquid SNEDDS for a certain time. SNEDDS tablets showed high shelf-life stability. In addition, compressed SNEDDS have shown faster in vitro dissolution rate and superior in vivo activity compared to conventional tablets. However, in some cases the bioavailability enhancement is lower than capsules and incomplete release from tablets was observed. Therefore, PWSD release should be monitored in bio-relevant media and the interactions between the SNEDDS and the adsorbents should be thoroughly evaluated.

REVIEW OF LITERATURE

2. REVIEW OF LITERATURE

1) **Akhilesh Dubey et al.** (2018) formulated and evaluated a novel solid self-nano emulsifying drug delivery system (SNEDDS) to increase the solubility and bioavailability of hydrochlorothiazide (HCZ). Enhancing both solubility and bioavailability of drugs remain the cornerstone for achieving successful outcomes of delivery systems. The most important is the protection of the drug from enzymatic or chemical degradation. Liquid SNEDDS (L-SNEDDS) was prepared by adding a drug to oil, surfactant, and co-surfactant and heated up to at 60°C under continuous stirring. Solid SNEDDS (S-SNEDDS) was prepared by mixing L-SNEDDS with microcrystalline cellulose in 1:1 proportion. Ex vivo skin permeation study indicated that 100% drug was released from both the L-SNEDDS and S-SNEDDS formulation SF3 in 3 h. Analysis of variance test showed significant differences (Moderately significant $P < 0.01$) in the values when compared to a marketed product. The prepared S-SNEDDS helped **in improving the solubility of the poorly soluble HCZ**, which is a step forward toward bioavailability enhancement and thus increased therapeutic efficacy of the drug[83].
2) **Tri Ujilestari et al**. (2018) formulated and characterized a self-nanoemulsifying drug delivery systems of cardamom (Amomum compactum) essential oil. The optimum formula was analyzed using a D-Optimal mixture designed by varying concentrations of oil component (Amomum compactum essential oil and virgin coconut oil), Tween 80, and polyethylene glycol 400 (PEG 400) (v/v) using a Design Expert® Ver. 7.1.5. Emulsification time and transmittance were selected as responses for optimization. SNEDDS of Amomum compactum essential oil was successfully formulated to SNEDDS using 10% of Amomum compactum essential oil, 10% of virgin coconut oil, 65.71% of Tween 80, and 14.29% of PEG 400. The characterization result showed the percent transmittance 99.37 ± 0.06, emulsification time 46.38 ± 0.61 s, the average droplet size 13.97 ± 0.31 nm with PI 0.06 ± 0.05, zeta potential −28.8 to −45.9 mV, viscosity 187.5 ± 0 mPa·s, passed the thermodynamic stress tests, and indicated spherical shape. The study revealed that the formulation has increased solubility and stability of Amomum compactum essential oil[84].
3) **Chuleegone Sornsuvit et al.** (2018) determined the pharmacokinetic parameters and bioavailability of **silymarin** 140mg SMEDDS formulation. An open-label, single-dose pharmacokinetic study was conducted. Twelve healthy volunteers were included in the study. After the volunteers had fasted overnight for 10 h, a single-dose generic silymarin 140mg SMEDDS soft capsule was administered. The pharmacokinetic parameters were calculated after silymarin had been administered as a single capsule. The mean (range) Cmax was 812.43 (259.47–1505.47) ng/ml at 0.80 (0.25–1.67) h (tmax). The mean (range) AUC0-t and AUC0-inf were658.80 (268.29–1045.01) ng.h/ml and 676.98 (274.10–1050.96) ng.h/ml,

respectively.Themean ke and t1/2 were 0.5386 h-1 and 1.91 h, respectively. The silymarin SMEDDS formulation soft capsule **showed rapid absorption and high oral bioavailability**[85].

4) **Chunxia Liu et al.** (2018) investigated the potential of self-nano emulsified drug delivery system (SNEDDS) to improve the oral bioavailability of **tetrandrine** (Tet). SNEDDS was developed by using rational blends of excipients with good solubilizing ability for Tet which was selected based on solubility studies. The optimal formulation with the best self-nano emulsified and solubilization ability consisted of 40% (w/w) oleic acid as oil, 15% (w/w) SPC and 30%(w/w) Cremophor RH-40 as surfactant, and 15%(w/w) PEG400 as co-surfactant. The dissolute rate of Tet SNEDDS in various dissolution media was remarkably faster than Tet commercial tablet. Moreover, in vivo pharmacokinetic study results show that significant increase ($p \leq 0.05$) in the peak concentration (Cmax) and the area under the curve (AUC) of Tet was observed after the oral administration of Tet SNEDDS and the absorption of Tet from SNEDDS resulted in approximately 2.33-fold increase in oral bioavailability compared with the commercial tablet. Our research suggests that the prepared Tet SNEDDS could be a good candidate for **improved the dissolution and oral bioavailability** of Tet[86].
5) **Gannu Praveen Kumar et al.** (2018) formulated, characterized and evaluated DFS entrapped SNEDDS. SNEDDS were prepared by emulsion diffusion evaporation technique. The drug released for 3-4 hrs approximately in all the media for 50% of release to occur except in SGF and FASGF in which the total percent release itself was 50% till 30 h. A significant reduction in toxicity of DFS was observed for SNEDDS in gastric mucosa when compared to free DFS with a 10 fold decrease in ulcer index of DFS in acute study and a 6.5 fold decrease in ulcer index of DFS in chronic study[87].
6) **Suvendu Kumar Sahoo et al.** (2018) developed a self-nano emulsifying drug delivery system (SNEDDS) for the oral delivery of **aripiprazole** (APZ), In this investigation, attempts were made to enhance the aqueous solubility of APZ through SNEDDS and to assess its effect on oral bioavailability (BA) in rabbit. Self-emulsifying drug delivery systems of APZ were formulated with Anise essential oil as oil phase, Gelucire 44/14 as surfactant, nd Transcutol HP as cosurfactant after screening various vehicles. The pharmacokinetic study in rabbits showed that SNEDDS has significantly increased the area under the curve[88].
7) **Fang Li et al.** (2017) developed a self- micro emulsifying drug delivery system (SMEDDS) of Hup-A to enhance the oral bioavailability and lymphatic uptake and transport of Hup-A. A single-pass intestinal perfusion (SPIP) technique and a chylomicron flow-blocking approach were used to study its intestinal absorption, mesenteric lymph node distribution and intestinal lymphatic uptake. The value of the area under the plasma concentration–time curve (AUC) of Hup-A SMEDDS was significantly higher than that of a Hup-A suspension (Po0.01). For Hup-A SMEDDS, the values of AUC and maximum plasma concentration (Cmax) of the

blocking model were significantly lower than those of the control model (Po0.05). The proportion of lymphatic transport of Hup-A SMEDDS and Hup-A suspension were about 40% and 5%, respectively, suggesting that SMEDDS can significantly **improve the intestinal lymphatic uptake and transport** of Hup-A[89].

8) **Liza Pratiwi et al.** (2017) formulated Self-Nanoemulsifying Drug Delivery System (SNEDDS) of **mangosteen peels** and evaluated the permeation ability of active substances in the formulation. The formulation was designed with a simplex lattice design using Design Expert software and the permeation was tested using Franz diffusion cell. Based on the results of simplex lattice design methods obtained that the optimum formulation of SNEDDS was the composition of virgin coconut oil (VCO), Tween 80, PEG 400 at a ratio of 1:6,95:2,05. The results of permeation test in vitro using Franz Diffusion cell indicated that the obtained SNEDDS ethyl acetate fraction of mangosteen peels that is 96.9223% higher than without preparation SNEDDS was 18,9426 % on hour-8. SNEDDS can improve the diffusion rate of mangosteen peels as a model poorly water soluble drug. Various samples of mangosteen peels were screened as candidates for SNEDDS on the basis of solubility of the active compound in oils, surfactants, and co-surfactants. Simplex lattice design methods can be used to obtain optimum formulation on SNEDDS[90].
9) **Ali naser et al.** (2016) optimized the different conditions for the preparation of selfnanoemulsifying drug delivery system (SNEDDS) for both **Irbesartan** (IRB) and Olmesartan (OLM). Sixteen unloaded SNEEDS formulae were prepared using Capryol 90, **Cremophor RH 40** and Transcutol HP as oil, surfactant and cosurfactant respectively and it was evaluated. It was concluded that the prepared self-emulsified prototype was ready to incorporate many poorly soluble drugs in order to **improve their solubility as well as bioavailability profile[91]**.
10) **Ahmed Alaa Kassem et al.** (2016) developed and optimized self-nanoemulsifying drug delivery systems (SNEDDSs) to improve the per-oral bioavailability of poorly soluble polyene antifungal drug, **Nystatin (NYS),** and to evaluate its in vitro and in vivo performance. Oleic acid (oil), Tween® 20 (Tw20) and Tween® 40 (Tw40) (surfactants) as well as dimethyl sulfoxide (DMSO) and propylene glycol (PG) (co-surfactants) were employed to construct pseudo-ternary phase diagrams. Five optimized formulations composed of oleic acid,Tw20 and DMSO or PG at Smix ratios (1:1, 2:1 or 3:1) were selected. They were spherical in shape of mean droplet size b100 nm with negatively charged zeta potential b−15mV. The in vitro release profile of NYS-SNEDDS was found significant in comparison to the plain NYS suspension. In vitro and in vivo evaluations against Candida albicans depicted promoted antifungal efficacy of selected NYS-SNEDDS formulations compared to marketed and plain NYS suspensions. The results indicate that NYS loaded

SNEDDS, with enhanced solubilization and nano sizing, has potential to **improve the absorption of drug and increase its oral antifungal efficacy[92].**

11) **Zhao et al.** (2016) demonstrated in vitro anti-cancer effects of silibinin. However, as many other drugs, silibinin can degrade in the stomach due to the action of the gastric fluid. The use of **pH-sensitive self-nanoemulsifying drug delivery systems (pH-SNEDDS)** could overcome the drawback due to degradation of the drug in the stomach while enhancing its solubility and dissolution rate. In vitro drug release studies of the optimal pH-SNEDDS indicated substantial increase of the drug release and release rate in comparison to pure silibinin and to the commercial silibinin tablet. The results indicated that pH-SNEDDS have potential to improve the bio-pharmaceutics properties of acid-labile lipophilic drugs[93].
12) **Dong Woo Yeom et al.** (2016) improved the dissolution and oral bioavailability (BA) of **atorvastatin calcium (ATV),** optimized self-microemulsifying drug delivery system (SMEDDS) using Capmul® MCM (oil), Tween® 20 (surfactant), and tetraglycol (cosurfactant). In this study, various solid carriers were employed to develop a solidified SMEDDS (S-SMEDDS): mannitol (M) and lactose (L) as water-soluble carriers, and Sylysia® 350 (S) and Aerosil® 200 (A) as water-insoluble carriers. S-SMEDDS with mannitol (S(M)-SMEDDS) or lactose (S(L)-SMEDDS) had a smaller droplet size and greater dissolution than S-SMEDDS with Sylysia® 350 (S(S)-SMEDDS) or Aerosil® 200 (S(A)-SMEDDS). Following oral administration of various formulations to rats at a dose equivalent to 25 mg/kg of ATV, plasma drug levels were measured by LC-MS/MS. The relative Bas (RBAs) of SMEDDS, S(M)-SMEDDS, and S(S)-SMEDDS were 345%, 216%, and 160%, respectively, compared to that of ATV suspension. Additionally, at a reduced dose of ATV equivalent to 5 mg/kg, the RBAs of S(M)-SMEDDS and S(S)-SMEDDS compared to that of SMEDDS were 101% and 65%, respectively. These results suggest that S(M)-SEMDDS offers great potential for the development of solid dosage forms with **improved oral absorption of drugs with poor water solubility**[94].
13) **Ali Nasr et al.** (2016) developed a solid self-nanoemulsifying drug delivery system (S-SNEDDS) of Olmesartan (OLM) for enhancement of its solubility and dissolution rate. In this study, liquid SNEDDS containing Olmesartan was formulated and further developed into a solid form by the spray drying technique using Aerosil 200 as a solid carrier. Based on the preliminary screening of different unloaded SNEDDS formulae, eight formulae of OLM loaded SNEEDS were prepared using Capryol 90, Cremophor RH40 and Transcutol HP as oil, surfactant and cosurfactant, respectively. In vitro release of OLM from SNEDDS formulae showed that more than 90% of OLM released in approximately 90 min. Optimized SNEDDS formulae were selected to be developed into S-SNEDDS using the spray drying technique. The prepared S-SNEDDS formulae were evaluated. To clarify the possible improvement in pharmacokinetic behavior of OLM S-SNEDDS, plasma concentration-time

curve profiles of OLM after the oral administration of optimized S-SNEDDS formula (F3) were compared to marketed product and pure drug in suspension. At all time points, it was observed that OLM plasma concentrations in rats treated with S-SNEDDS were significantly higher than those treated with the drug in suspension and marketed product[95].

14) **Erna Wulandari et al.** (2016) formulated self-nanoemulsifying drug delivery System (SNEDDS) of β -carotene to facilitate oral delivery. SNEDDS of β -carotene consisted of olive oil as the oil phase, Tween 80 as the surfactant and PEG 400 as the co-surfactant. Optimisation of the composition was achieved with simplex lattice design (SLD) method, **software Design Expert version 7.1.5.** Optimum formula was determined by observing emulsification time and clarity of the nanoemulsions. Characterisation included physical stability in artificial gastric fluid (AGF), size and size distribution of the droplets and zeta potential value[96].

15) **Rajendra Narayan Dash et al.** (2015) explored a solid self-nanoemulsifying drug delivery system (solid SNEDDS) to improve the solubility and dissolution profile of **glipizide.** The optimized SNEDDS preconcentrate consisted of Captex 355 (30% w/w) as oil, Solutol HS15 (45% w/w) as surfactant and Imwitor 988 (25% w/w) as co-surfactant. The saturation solubility (SS) of glipizide in optimized SNEDDS preconcentrate was found to be 45.12± 1.36 mg/ml, indicating an improvement (1367 times) of glipizide solubility as compared to its aqueous solubility (0.033± 0.0021 mg/ml). At 90% SS, glipizide was loaded to the optimized SNEDDS. **Glipizide dissolution improved significantly** ($p < 0.001$) from the solid SNEDDS (100% in 15 min) as compared to the pure drug (18.37%) and commercial product (65.82) respectively[97].

16) **Spandana Inugala et al.** (2015) investigated the potential of solid self-nano emulsifying drug delivery system (S-SNEDDS) composed of capmul MCM C8 (oil), tween 80 (surfactant) and transcutol P (co-surfactant) in improving the dissolution and oral bioavailability of **darunavir**. In vitro drug release studies showed initial rapid release of about 13.3±1.4% within 30 min from L-SNEDDS followed by slow continuous release of entrapped drug and reached a maximum of 62.6±3.5% release at the end of 24h. In vitro dissolution studies indicated faster dissolution of darunavir from the developed S-SNEDDS with **3 times greater mean dissolution rate (MDR)** compared to pure darunavir. Furthermore, in vivo pharmacokinetic studies in Wistar rats resulted in enhanced values of peak drug concentration (Cmax) for L-SNEDDS (2.98±0.19 μg/mL) and S-SNEDDS (3.7±0.28μg/mL) compared pure darunavir (1.57±0.17 μg/mL)[98].

17) **Komal Parmar et al.** (2015) developed self-nanoemulsifying drug delivery system (S-SNEDDS) containing Capryol-90 as oil phase for the delivery of Embelin, a poorly water soluble herbal active ingredient. Box-Behnken experimental design was employed to optimise the formulation variables, X1 (amount of oil; Capryol 90), X2 (amount of surfactant; Acrysol

EL 135) and X3 (amount of co-surfactant; PEG 400). Optimised liquid formulations were formulated into free flowing granules (S-SNEDDS) by adsorption on the porous materials like Aerosil 200 and Neusilin and thereby compressed into tablet. In vitro dissolution studies of SNEDDS revealed increased in the dissolution rate of the drug Thus, the present studies demonstrated dissolution enhancement potential of porous carrier based S-SNEDDS for poorly water soluble herbal active ingredient, Embelin[99].

18) **Abdul Wadood Khan et al.** (2014) reported that **Naringenin** (NRG) predominant flavanone in grapefruits, possesses anti-inflammatory, anti-carcinogenic, hepato-protective and anti-lipid peroxidation effects. Slow dissolution after oral ingestion due to its poor solubility in water, as well as low bioavailability following oral administration, restricts its therapeutic application. Pseudo ternary phase diagrams were constructed to identify the area of nano emulsification. The developed self nano emulsifying drug delivery systems (SNEDDS) were evaluated. In vitro drug release from SNEDDS was significantly higher (p50.005) than pure drug. Furthermore, area under the drug concentration time-curve (AUC0–24) of NRG from SNEDDS formulation revealed a significant increase (p50.005) in NRG absorption compared to NRG alone. The increase in drug release and bioavailability as compared to drug suspension from SNEDDS formulation may be attributed to the **nano sized droplets and enhanced solubility of NRG** in the SNEDDS[100].

19) **Jyotsana R. Madan et al.** (2014) formulated liquid SMEDDS of **pioglitazone HCl** with Capmul MCM C8 and oleic acid as oil phase, **Cremophor RH 40** and Tween 80 as surfactant phase, and Transcutol P as cosurfactant phase after screening various vehicles. The optimized system possessed a mean globule size of 122.2 nm, zeta potential around -22.9 mV, drug content 99.66 ± 0.47%, viscosity 0.8874 ± 0.026 cP, emulsification time 38 s, polydispersity index value of 0.5, and transmittance value of 99.3 ± 0.6%. Drug release in hydrochloric acid buffer pH 2 was found to be 99.35 ± 0.38%. More than three-fold increase in dissolution characteristics of pioglitazone HCl in SMEDDS was observed as compared to pure and marketed formulation. Stability studies show there was no sign of phase separation or precipitation and no change in drug content was observed[101].

20) **Rahul Shankar Narkhede et al.** (2014) formulated self-nano emulsifying drug delivery of nebivolol hydrochloride (NEB) to increase the bioavailability of drug by increasing solubility and permeability through the gastro intestinal membrane. The optimum concentration of a system determined Capmul MCM EP 25% as oil, Tween-60 50% as surfactant, Transcutol HP 12.5%, PEG-400 12.5% as co-surfactant, with a globule size of 124.5 nm, cloud point at 770C and zeta potential of -5.123 mV. In-vitro drug release study and ex-vivo permeation study showed significant **increase in dissolution rate and permeability** respectively, as compared to the drug suspension and marketed preparation (NEBISTAR™)[102].

21) **Shailesh T. Prajapati et al.** (2013) concluded that SMEDDS would be a **promising drug delivery system for poorly water-soluble drugs by the oral route**. Olmesartan medoxomil (OLM) is an angiotensin II receptor blocker (ARB) antihypertensive agent administered orally that has absolute bioavailability of only 26% due to the poor aqueous solubility (7.75 g/ml). Pseudoternary phase diagrams were constructed using Acrysol EL 135, Tween 80, Transcutol P, and distilled water to identify the efficient self-microemulsification region. The optimized formulation S2 contained OLM (20 mg), Tween 80 (33%v/v), Transcutol P (33%v/v), and Acrysol EL 135 (34%v/v) had shown the smallest particle size, maximum solubility, less emulsification time, good optical clarity, and in vitro release. The in vitro and ex vivo diffusion **rate of the drug from the SMEDDS was significantly higher than that of the plain drug suspension**[103].

AIM AND PLAN OF WORK

3. AIM AND PLAN OF WORK

The main aim of this work is to prepare SMEDDS for oral solubility and bioavailability enhancement of poorly water soluble drug. SMEDDS spread readily in the GI tract, and the digestive motility of the stomach and the intestine provides the agitation necessary for self-emulsification. When compared with emulsions which are sensitive and Meta stable dispersed forms, SMEDDS are physically stable formulations that are easy to manufacture. Thus, for lipophilic drug compounds which 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.

This study is focused to formulate the solid self micro emulsifying drug delivery system of Ivermectin for improvement of solubility by providing large interfacial surface area and thereby increasing the bioavailability of Ivermectin used in parasitic infections.

Self micro emulsifying drug delivery system is generally used to improve the solubility of the poorly water soluble drug and thereby improve the bioavailability of drugs. They provide large interfacial surface area by reducing the size of the particles. Solid SMEDDS generally improve the patient compliance than the liquid SMEDDS. By improving the solubility and bioavailability, we can reduce the dosing frequency and drug loading in single unit dosage form.

In the present research work, we have attempted to develop solid self-micro emulsifying drug delivery system. As SMEDDS provides large surface area due to small globule size, they improve the solubility and bioavailability of Ivermectin.

The solid self-micro emulsifying drug delivery system of Ivermectin will have the following advantages:

- Improve storage stability
- Improve oral bioavailability
- Increased drug loading capacity
- Reduces inter- and intra-subject variability in drug pharmacokinetics
- Reduce production cost and simplifies manufacturing of products

Part I

- Determination of λmax
- Drug-Excipient compatibility study by FT-IR

Part II

- Calibration of ivermectin in ethanol solution .
- Intrinsic Dissolution study of Ivermectin.

Part III

- Determining solubility of Ivermectin and screening of components.
- Construction of ternary phase diagram.
- Optimization by using D- Optimal design.

Part IV

- Preparation of L-SMEDDS formulations.
- Evaluations of L-SMEDDS formulations.
    - ✓ Self-emulsification time.
    - ✓ % Transmittance.
    - ✓ Refractive index.
    - ✓ pH of L-SMEDDS.
    - ✓ Viscosity of L-SMEDDS.
    - ✓ Drug content of L-SMEDDS.
    - ✓ Dispersibility test.
    - ✓ Globule size and zeta potential analysis.
    - ✓ In vitro Drug release study of L-SMEDDS.

RATIONALE OF THE STUDY

4. RATIONALE OF THE STUDY

Oral route still remains the favourite route of drug administration in many diseases and till today it is the first way investigated in the development of new dosage forms. The major problem in oral drug formulations is low and erratic bioavailability, which mainly results from poor aqueous solubility. This may lead to high inter- and intra subject variability, lack of dose proportionality and therapeutic failure[104, 105].

It is estimated that 40% of active substances are poorly soluble in water. The improvement of bio-availability of drugs with such properties presents one of the greatest challenges in drug formulations. Various technological strategies are reported in the literature including micronization, solid dispersions or cyclodextrines complex formation and different technologies of drug delivery systems. Among various approach self nano emulsifying drug delivery system (SNEDDS) has gained more attention due to enhanced oral bio-availability enabling reduction in dose [12.]

SNEDDS are isotropic mixtures of oils, surfactants, co-solvents and can be used for the design of formulations in order to improve the oral absorption of highly lipophilic compounds. SNEDDS emulsify spontaneously to produce fine oil-in-water emulsions when introduced into GI fluid under gentle agitation (GI motility).

SNEDDS 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 plasma level concentrations.

Stimulation of body secretions help in digestion of lipids: Administration of lipid can stimulate the biliary and pancreatic secretions which are helpful for the digestion of lipids. The enzymes present in the secretions are water soluble and act at water/lipid interface Prolongation of GI residence time: administration of lipid along with the drug allows the drug to be present for prolonged duration of period in the GIT which facilitates the absorption of the drug[2].

Stimulation of lymphatic transport: the highly lipophilic drug ($\log P > 5$) which has high solubility in triglycerides (>50 mg/mL) can undergo lymphatic transport when co-administered with esters of unsaturated long chain fatty acids; thereby bioavailability

Can be improved [7–9]. This restricted lymphatic transport is mainly due to low lymph to-blood flow ratio. This enhanced lymph delivery of the drug can bypass the first pass extraction whereby the bioavailability of drugs that undergo extensive first pass effect can be improved. Increased intestinal wall permeability: opening of tight junctions in the intestine caused by lipids contributes to the increased permeability of poorly permeable drugs[7]. Although this mechanism is not essential in case of BCS Class II drugs, it leads to marked improvement in absorption of Class IV drugs which have both dissolution and permeability rate limited absorption.

Reduced efflux of the drug in the GIT: lipids such as anionic phospholipids (cardiolipin and phosphatidylserine) may inhibit permeability glycoprotein (P-gp) by interaction with membrane lipids. So the drugs which have propensity to be effluxes from the GIT can be formulated as lipid based delivery systems for the improvement of bioavailability11. The inhibitory effect is due to competition for binding with the transporter and due to membrane perturbation caused by the excipients, mainly surfactants. The residence time of the drug can be prolonged by this inhibition of efflux[8].

Among various lipid based formulations (liposome's, solid lipid nanoparticles, self-dispersing tablets, and solid solutions), self nanoemulsifying formulations are receiving more attention by formulation scientists as these are advantageous in the aspect of their stability, self-dispersing nature, ease of preparation, and scale-up. SNEDDS are the isotropic, clear mixtures of oils and surfactants and sometimes include co solvents/co surfactants. These are designed to form o/w nanoemulsions with mild agitation produced by the motility of GIT followed by solubilisation and absorption of drug. SNEDDS usually produce nanoemulsions of droplet size below 100 nm upon dilution[3].

DISEASE PROFILE

5. DISEASE PROFILE

Onchocerciasis[115]:

- Onchocerciasis, commonly known as "river blindness", is caused by the parasitic worm Onchocerca volvulus.
- It is transmitted to humans through exposure to repeated bites of infected black flies of the genus Simulium
- Symptoms include severe itching, disfiguring skin conditions, and visual impairment, including permanent blindness.
- More than 99% of infected people live in 31 African countries. The disease also exists in some foci in Latin America and Yemen.
- The Global Burden of Disease Study estimated in 2017 that there were 20.9 million prevalent O. volvulus infections worldwide: 14.6 million of the infected people had skin disease and 1.15 million had vision loss

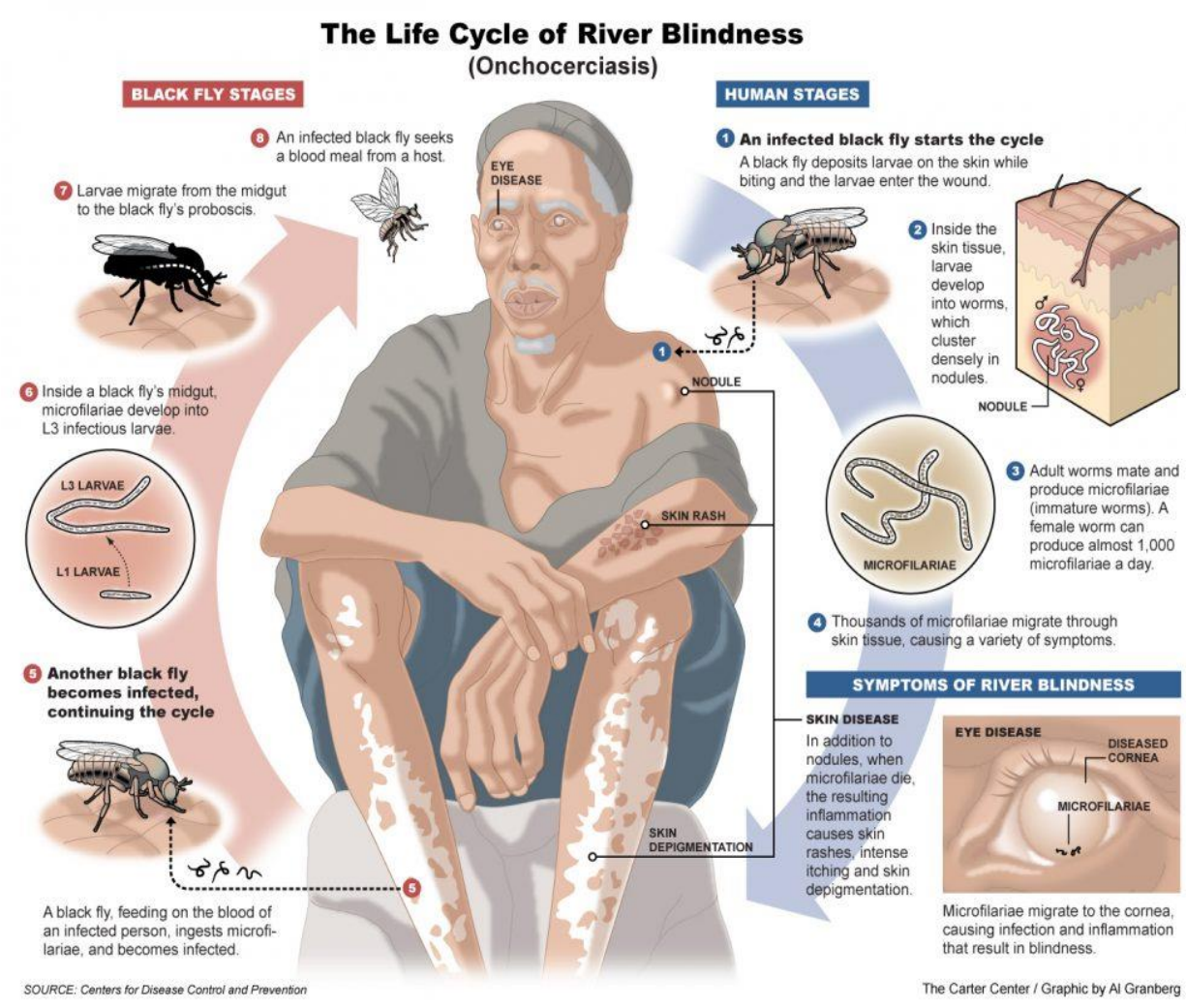


Fig.3 Life cycle of river blindness.

Ascariasis[113]:

An estimated 807 million–1.2 billion people in the world are infected with *Ascaris lumbricoides*. *Ascaris*, hookworm, and whipworm are parasitic worms known as soil-transmitted helminths (STH). Together, they account for a major burden of parasitic disease worldwide.

Ascaris parasites live in the intestine. *Ascaris* eggs are passed in the faeces of infected people. People with ascariasis often show no symptoms. If symptoms occur they can be light. Symptoms include abdominal discomfort or pain. Heavy infections can block the intestines and slow growth in children.

Humans can also be infected by pig roundworm (*Ascaris suum*). *Ascaris lumbricoides* (human roundworm) and *Ascaris suum* (pig roundworm) are hard to tell apart.

Fig.4 Ascariasis Left/Right: Fertilized eggs of *A. lumbricoides* in unstained wet mounts of stool. Centre: Adult female *A. lumbricoides*.

STRONGYLOIDIASIS[114]:

Strongyloidiasis is a disease caused by a nematode, or a roundworm, in the genus *Strongyloides*. Though there are over 40 species within this genus that can infect birds, reptiles, amphibians, livestock and other primates, *Strongyloides stercoralis* is the primary species that accounts for human disease.

The majority of people infected with *Strongyloides* do not have symptoms. Those who do develop symptoms often have non-specific, or generalized complaints. Some people develop abdominal pain, bloating, heartburn, intermittent episodes of diarrhoea and constipation, a dry cough, and skin rashes. Rarely people will develop arthritis, kidney problems, and heart conditions.

Strongyloidiasis can be severe and life-threatening in persons who

- Are taking corticosteroids (oral or intravenous) for asthma or chronic obstructive pulmonary disease (COPD) exacerbations, lupus, gout, or other conditions requiring steroids for immunosuppressant or symptomatic relief;
- Are infected with the virus HTLV-1;

- Have hematologic malignancies such as leukaemia or lymphoma; or
- Are transplant recipients.

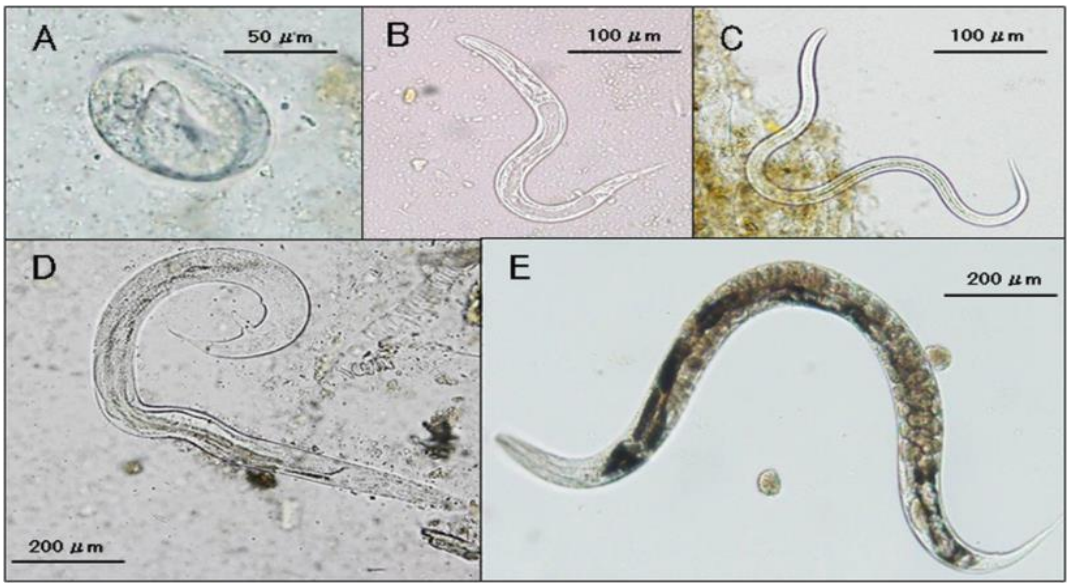


Fig. 5 various growth patterns of *Strongyloides stercoralis* observed in patient faeces by a microscope. (**A**) Embryonated egg, 80 μm × 40 μm. (**B**) Rhabditiform larvae, 300 μm × 18 μm. (**C**) Filariform larvae, 500 μm × 15 μm. (**D**) Adult male of free-living stage, 1 mm × 50 μm. (**E**) Adult female of free-living stage, 1.2 mm × 70 μm.

DRUG PROFILE

6. DRUG PROFILE

Ivermectin:

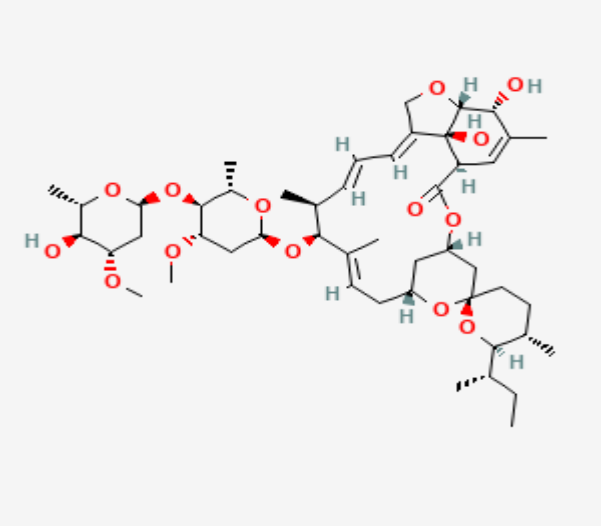
Ivermectin is a semi synthetic derivative of avermectin B_1 and consists of an 80:20 mixture of the equipotent homologous 22,23 dehydro B_{1a} and B_{1b}. Ivermectin works through many mechanisms of action that result in the death of the targeted parasites it can be taken by mouth or applied to the skin for external infestations. Parasitic infestations in humans include head lice, scabies, onchocerciasis, strongyloidiasis, trichuriasis, and ascariasis.

Category:

The drug belongs to the avermectin family of medications.

Physiochemical properties:

Chemical structure[111]:



IUPAC name **:** (1'R, 2R, 4'S, 10'E, 14'E, 16'E, 21'R)-6-(butan-2-yl)-21', 24'-dihydroxy-12'

- {[(2R, 4S, 6S)-5-{[(2S, 4S, 6S) - 5-hydroxy-4-methoxy-6-methyloxan-2-yl] oxy}-4-methoxy-6-methyloxan-2-yl] oxy}-5, 11', 13', 22'

-Tetramethyl-3', 7', 19'-trioxaspiro [oxane-2, 6'- tetracyclo [15.6.1.1^ {4, 8}.0^ {20, 24}] pentacosane]-10', 14', 16', 22'-tetraen-2'-one.

CAS number **:** 70288-86-7

Description **:** A white crystalline powder

Molecular weight **:** 875.1 g/mol

Molecular formula **:** $C_{48}H_{74}O_{14}$

Melting point **:** 155°C

Solubility **:** It is insoluble in water but is freely soluble in methanol and Soluble in 95% ethanol.

Log P value **:** 4.1

Pharmacokinetics[106, 107]:

BCS classification **:** BCS class Ⅱ drug (low solubility, high permeability)

Absorption: Ivermectin has a low solubility in water. Its absorption from water-free formulations is slower than from water-micelle solutions and its biological half-life is longer. The longer residence of ivermectin resulting from water-free injection formulations is reflected in the prolonged duration of its clinical efficacy. With oral formulations, the bioavailability of ivermectin is 20 % greater when using a micelle solution (applied through a nasogastric tube) compared to an oral paste. Ivermectin's disposition is strongly dependent on its route of administration. Significant differences, in its bioavailability and biological half-life, between an oral and a subcutaneous administration.

Half-life: 12-36 hrs.

Volume of distribution: Ivermectin is highly lipophilic, and distributes widely in the body with a volume of distribution (V_d) of 3.1–3.5 l/kg.

Protein binding: Studies suggest that Ivermectin is a highly protein bound drug shows a great degree of protein binding (>90%).

Metabolism: Studies regarding the metabolism of ivermectin in humans are scarce. This drug is extensively metabolized by human liver microsomes by cytochrome P450. The predominant isoform responsible for the biotransformation of this compound in the liver of humans is cytochrome P-4503A4, converting the drug to at least 10 metabolites, most of them hydroxylated and demethylated derivatives.

Elimination: Ivermectin and its metabolites were excreted mainly in faeces and only 1% in urine. Positive identification was obtained for the presence of 3"-*O*-desmethyl-H_2B_{1a}, and 22, 23-dihydroavermectin B_{1a} monosaccharide in urine and faeces, respectively.

Pharmacodynamics[107]:

Ivermectin and its related drugs act by **interfering with the nerve and muscle functions of helminths and insects**. The drug binds to glutamate-gated chloride channels common to invertebrate nerve and muscle cells.

Mechanism of action:

Ivermectin and its related drugs act by interfering with the nerve and muscle functions of helminths and insects. The drug binds to **glutamate-gated chloride channels common to**

invertebrate nerve and muscle cells. The binding pushes the channels open, which increases the flow of chloride ions and hyper-polarizes the cell membranes, paralyzing and killing the invertebrate. Ivermectin is safe for mammals (at the normal therapeutic doses used to cure parasite infections) because mammalian glutamate-gated chloride channels only occur in the brain and spinal cord: the causative avermectins usually do not cross the blood–brain barrier, and are unlikely to bind to other mammalian ligand-gated channels.

Therapeutic uses:

Ivermectin is an broad spectrum anthelmintic mainly used for parasitic infections in humans and animals

It acts by interfering with the nerve and muscle functions of helminths and insects. The drug binds to glutamate-gated chloride channels common to invertebrate nerve and muscle cells.

- River blindness (onchocerciasis)
- Intestinal strongyloidiasis
- Trichuriasis
- Ascariasis
- Rosacea

Dosage:

- For river blindness:
    - Adults and teenagers—Dose is based on body weight and must be determined by your doctor. The usual dose is 150 micrograms (mcg) per kilogram (kg) (68 mcg per pound) of body weight as a single dose. The treatment may be repeated every three to twelve months.
    - Children—Dose is based on body weight and must be determined by your doctor. For children weighing 15 kg (33 pounds) or more, the usual dose is 150 mcg per kg (68 mcg per pound) of body weight as a single dose. If necessary, the treatment may be repeated every three to twelve months. For children weighing less than 15 kg, use and dose must be determined by your doctor.

Drug interactions[109]:

Studies show that ivermectin causes serious life threatening interaction with these drugs

- Erdafitinib (erdafitinib will increase the level or effect of ivermectin by P-glycoprotein (MDR1) efflux transporter.)
- Lasmiditan (lasmiditan increases levels of ivermectin by P-glycoprotein (MDR1) efflux transporter.)

- Quinidine (increases levels of ivermectin by P-glycoprotein (MDR1) efflux transporter.)
- Sotorasib (increases levels of ivermectin by P-glycoprotein (MDR1) efflux transporter.)
- Tepotinib (increases levels of ivermectin by P-glycoprotein (MDR1) efflux transporter.)

Adverse drug reaction[108]:

Clinical trials and observational studies have reported common adverse events such as

- Headache
- Pruritus
- muscle pain
- Cough
- Dyspnea
- Nausea
- Vomiting
- Diarrhoea
- blurred vision
- postural hypotension and confusion and more anecdotal effects such as serious skin reactions and edematous swelling

EXCIPIENT PROFILE

7. EXCIPIENTS PROFILE

COCONUT OIL[121]

Non-proprietary Names

BP: Coconut Oil

JP: Coconut Oil

PhEur: Coconut Oil, Refined

USP-NF: Coconut Oil

Synonym

Aceite de coco; cocois oleum raffinatum; coconut butter; copra oil; oleum cocois; pureco 76; refined coconut oil.

Chemical Name and CAS Registry Number

Coconut oil [8001-31-8]

Empirical Formula

Coconut oil contains triglycerides, the fatty acid constituent of which are mainly lauric and myristic acids with smaller proportions of capric, caproic, oleic, palmitic and stearic acids.

The PhEur 6.2 and USP32-NF27 state that the fatty acid composition for coconut oil is caproic acid (≤1.5%), Caprylic acid (5-11%), Capric acid (4-9%), Lauric acid (40-50%), Myristic acid (15-20%), Palmitic acid (7-12%), Stearic acid (1.5-5%), Arachidic acid (≤0.2%), Oleic acid (4-10%), Linoleic acid (≤0.2%) and Eicosenoic acid (≤0.2%).

Functional Category

Oleaginous vehicle, Emollient

Description

Coconut oil is generally occurs as a white to light yellow mass or colourless or light-yellow clear oil, with a slight odour characteristic of coconut oil and mild taste. Refined coconut oil is a white or almost white unctuous mass.

The form that coconut oil takes depends on temperature; it occurs as pale yellow to colourless liquid between 28ºC, and as a hard brittle crystalline solid below15ºC.

Typical Properties

Flash point: 216ºC (closed up)

Melting point: 23-26ºC

Boling point: >450ºC

Specific gravity: 0.918-0.923

Refractive index: 1.448-1.450

Saponification number: 255-258 (mg KOH/g)

Iodine value: 8-9.5

Solubility

Practically insoluble in water, freely soluble in dichloromethane and in light petroleum (bp: 65-70ºC); soluble in carbon di sulfide, chloroform and ether; soluble at 60ºC in 2 parts of ethanol (95%) but less soluble at lower temperatures.

Surface tension

33.4 mN/m (dynes/cm) at 20oC; 28.4 mN/m (dynes/cm) at 80ºC.

Application

- Coconut oil has traditionally been used in ointments where it forms a readily absorbable base. It has been used particularly in preparations intended for application to the scalp, where it could be applied as a solid but would liquefy when applied to the skin.

- Coconut oil is readily saponified by strong alkalis even in the cold and as the soap produced is not readily precipitated by sodium chloride, it has been used in the making of 'marine' soap.
- It may be used in the formulation of a range of other preparation including emulsions, nanoemulsion, intranasal solution, rectal capsules and suppositories.
- In addition, coconut oil has been reported to have antifungal activity against a range of Candida species.
- It has been used therapeutically in a lotion for the eradication of head lice, and was included in regime used to treat a patient who had ingested 16.8 g aluminium phosphide.
- Concern has been expressed at the potential use of coconut oil as a suntan lotion as it does not afford any protection against UV light.

Stability and Storage Conditions

Coconut oil remains edible, and mild in taste and odour, for several years under ordinary conditions. However, on exposure to air, the oil readily oxidizes and becomes rancid, acquiring an unpleasant odour and strong acid taste.

Store in tight, well-filled container, protected from light at a temperature not exceeding 25ºC. Coconut oil may be combustible at high temperature, and may spontaneously heat and ignite if stored under hot and wet conditions.

OLIVE OIL[122]

Non-proprietary Names

BP: Refined Olive Oil

JP: olive Oil

PhEur: Olive Oil, Refined

USP-NF: Olive Oil

Synonym

Gomenoleo oil, olivae oleum raffinatum, pure olive oil, olea euro pea oil, oleum olivae

CAS Registry Number

8001-25-00

Empirical Formula

Olive oil is a mixture of fatty acid glycerides. Analysis of olive oil shows a high proportion of unsaturated fatty acids.

Functional Category

Oleaginous vehicle

Description

Olive oil is the fixed oil obtained by cold expression or other suitable mechanical means from the ripe drupes of olea europaea. It occurs as a clear, colourless or yellow, transparent oily liquid. It may contain suitable antioxidants.

Typical Properties

Specific gravity: 0.908-0.914 g/cm3

Refractive index: 1.4657-1.4893

Saponification value: 186-194 (mg KOH/g)

Acid value: ≤ 1.0(mg KOH/g)

Iodine value: 79-88

Solubility

Slightly soluble in ethanol (95%), miscible with ether, chloroform, light petroleum and carbon di sulfide.

Application

Olive oil has been used in enemas, liniments, ointments, plasters and soap. It has also been used in oral capsules and solutions, and as a vehicle for oily injections including targeted drug delivery system.

In cosmetics, olive oil used as a solvent, and also a skin and hair conditioner.

Stability and Storage

When cooled, olive oil becomes cloudy at approximately 100C, and becomes butter like mass at 0^{o} c.

Olive oil should be stored in a cool, dry place in a tight, well filled container, protected from light.

CASTOR OIL[123]

Non-proprietary Names

BP: Virgin Castor oil

JP: Castor Oil

PhEur: Castor Oil, virgin

USP-NF: Castor Oil

Synonym

Oleum ricini, ricini oleum virginale, ricinoleum, ricinus communis, ricinus oil, Lipovol CO.

CAS Registry Number

8001-79-4

Empirical Formula

Castor oil is a triglyceride of fatty acids. The fatty acid composition is approximately ricinoleic acid (87%); oleic acid (7%); linoleic acid (3%); palmitic acid (2%); stearic acid (1%) and trace amounts of dihydroxystearic acid.

Functional Category

Emollient; oleaginous vehicle; solvent.

Description

It is a clear, almost colourless or pale yellow colored viscous oil. It has a slight odour and taste that is bland initially but afterwards slightly acrid.

Typical Properties

Specific gravity: 0.953-0.965

Refractive index: 1.473-1.477

Viscosity: 1000 cP at 20ºC and 200cP at 40ºC

Saponification value: 176-187 (mg KOH/g)

Acid value: ≤ 1.5(mg KOH/g)

Iodine value: 80-90

Solubility

Miscible with chloroform, diethyl ether, ethanol, glacial acetic acid and methanol. Freely soluble in ethanol (95%) and petroleum ether. Practically insoluble in water.

Application

- Castor oil widely used in cosmetics, food products and pharmaceutical formulations.
- In pharmaceutical formulations castor oil is most commonly used in topical creams and ointments at concentration of 5-12.5 %.
- It also used in oral tablet and capsule formulations, ophthalmic emulsions and as a solvent in intramuscular injections.

Stability and Storage conditions

Castor oil is stable and does not turn rancid unless subjected to excessive heat. On heating at 3000C for several hours, castor oil polymerizes and becomes soluble in mineral oil. When cooled to 00C, it becomes more viscous.

Castor oil should be stored at a temperature not exceeding 250C in a well filled airtight containers protected from light

ROSE OIL[124]

Non-proprietary Names

BP: Rose oil

JP: Rose Oil

PhEur: Rose Oil,

USP-NF: Rose Oil

Synonym

Attar of roses, Otto of rose, essence of rose, rose oil

CAS Registry Number

8007-01-0

Empirical Formula

The main compounds of Rose oils were as follows: β-citronellal (30.24–31.15%); trans-geraniol (20.62– 21.24%), n-heneicosane (8.79–9.05%), n-nonadecane (8.51– 8.77%), nonadecene (4.42–4.55%) and phenyl ethyl alcohol (4.04–4.16%).

Functional Category

Fragrance; oleaginous vehicle; solvent.

Description

Rose oil is a fragrant, colourless or pale-yellow liquid essential oil distilled from fresh petals of Rosa damascene and R. gallica and other species of the rose family Rosaceae.

Typical Properties

Specific gravity: 0.84800 - 0.86100 at 25°C

Refractive index: 1.45300 - 1.46400 at 20°C

Relative density: 0.950 to 0.995 g/cm3

Ester value: 13- 30 (mg KOH/g)

Acid value: ≤ 11.0 (mg KOH/g)

Iodine value: 152-169

Solubility

Miscible with chloroform, diethyl ether, ethanol, glacial acetic acid and methanol. Freely soluble in ethanol (95%) and petroleum ether. Practically insoluble in water.

Application

- Rose oil soothes and harmonizes the mind and helps with depression, anger, grief, fear, nervous tension and stress and at the same time addresses sexuality, self-nurturing, self esteem and dealing with emotional problems.
- It is very helpful for poor circulation and heart problems, which includes heart palpitations, arrhythmia and high blood pressure as well.
- It is used in flavouring agent for cakes, candies, tea, jams, rose vinegars, salads and cream caramel. It is also used as additive to drinks, beverages and yogurt.
- It is mostly used in perfumery and also in toilet preparations, lozenges and toothpaste.

Stability and Storage Conditions

Rose oil should be stored at a temperature not exceeding 250C in a well filled airtight containers protected from light.

POLYSORBATE 80[125]

Non-proprietary Names

BP: Polysorbate 80

JP: Polysorbate 80

PhEur: Polysorbate 80

USP-NF: Polysorbate 80

Synonym

Tween 80, Emulgin SMO, polyoxyethylene 20 oleate, polysorbatum 80, Tego SMO 80.

Chemical Name

Polyoxyethylene 20 sorbitan monooleate

CAS Registry Number

9005-65-6

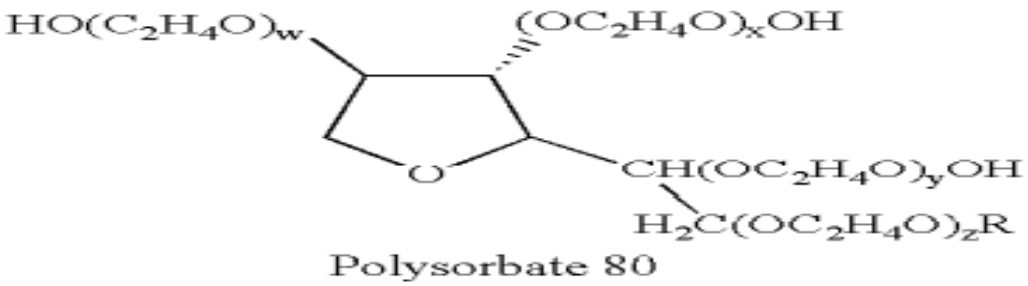
Empirical Formula

C64H124O26

Molecular Weight

1310

Structure



Polysorbate 80

Functional Category

Dispersing agent, emulsifying agent, Non-ionic surfactant, solubilising agent, suspending agent and wetting agent

Description

Polysorbates have a characteristic odour and a warm, somewhat bitter taste. It is a yellow oily liquid.

Typical Properties

Specific gravity: 1.08

Viscosity: 425 (mPas)

Saponification value: 45-55 (mg KOH/g)

Acid value: ≤ 2.0(mg KOH/g)

HLB value: 15.0

Hydroxyl value: 65-80

Solubility

It is soluble in water and ethanol. It is insoluble in mineral oil and vegetable oil.

Applications

- It is non-ionic surfactant widely used as emulsifying agent in the preparation of stable oil-in-water pharmaceutical emulsions.
- May also be used as solubilising agent for a variety of substance including essential oils and oil soluble vitamins.
- As wetting agent in the formulation of oral and parenteral suspensions.
- It also widely used in cosmetics and food products.

Stability and Storage Conditions

Polysorbates are stable to electrolytes and weak acids and bases. The oleic acid esters are sensitive to oxidation.

Polysorbates are hygroscopic. It should be stored in a well closed container, protected from light, in a cool and dry place.

POLYSORBATE 20[125]

Non-proprietary Names

BP: Polysorbate 20

JP: Polysorbate 20

PhEur: Polysorbate 20

USP-NF: Polysorbate 20

Synonym

Tween 20, polyoxyethylene 20 laurate, polysorbatum 20, capmul POE-L, sorbitan monododecanoate.

Chemical Name

Polyoxyethylene 20 sorbitan monolaurate

CAS Registry Number

9005-64-5

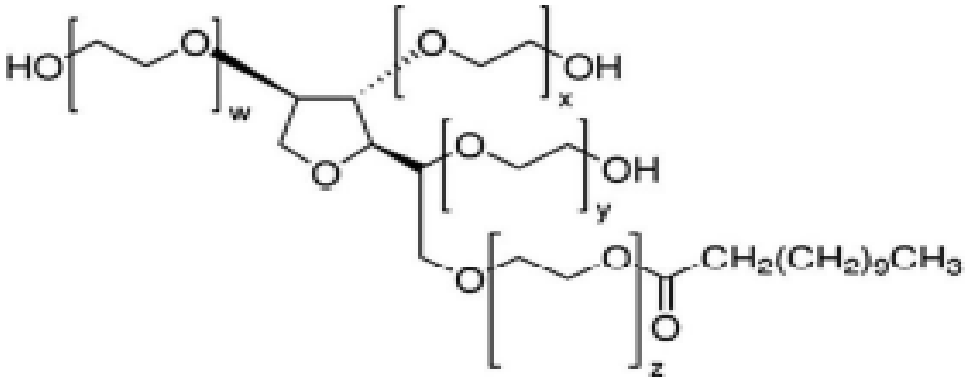
Empirical Formula

C58H114O26

Molecular Weight

1128

Structure



Functional Category

Dispersing agent, emulsifying agent, Non-ionic surfactant, solubilising agent, suspending agent and wetting agent

Description

Polysorbates have a characteristic odour and a warm, somewhat bitter taste. It is a yellow oily liquid.

Typical Properties

Specific gravity: 1.1

Viscosity: 400 (mPas)

Saponification value: 40-50 (mg KOH/g)

Acid value: ≤ 2.0(mg KOH/g)

HLB value: 16.7

Hydroxyl value: 96-108

Moisture content: 3.0 %

Solubility

It is soluble in water and ethanol. It is insoluble in mineral oil and vegetable oil.

Applications

- It is non-ionic surfactant widely used as emulsifying agent in the preparation of stable oil-in-water pharmaceutical emulsions.
- May also be used as solubilising agent for a variety of substance including essential oils and oil soluble vitamins.
- As wetting agent in the formulation of oral and parenteral suspensions.

Stability and Storage Conditions

Polysorbates are stable to electrolytes and weak acids and bases. The oleic acid esters are sensitive to oxidation.

Polysorbates are hygroscopic. It should be stored in a well closed container, protected from light, in a cool and dry place.

POLYSORBATE 20[125]

Non-proprietary Names

BP: Polysorbate 60

JP: Polysorbate 60

PhEur: Polysorbate 60

USP-NF: Polysorbate 60

Synonym

Tween 60, polyoxyethylene 60 laurate, polysorbatum 60, sorbitan monododecanoate.

Chemical Name

Polyoxyethylene 60 sorbitan dodecanoate

CAS Registry Number

9005-67-8

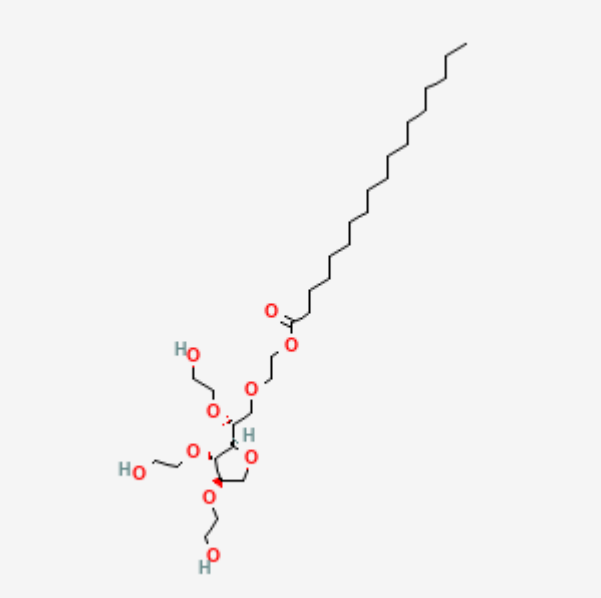
Empirical Formula

$C_{64}H_{126}O_{26}$

Molecular Weight

1311.7

Structure



Functional Category

Dispersing agent, emulsifying agent, Non-ionic surfactant, solubilising agent, suspending agent and wetting agent

Description

Polysorbates have a characteristic odour and a warm, somewhat bitter taste. It is a yellow oily liquid.

Typical Properties

Specific gravity: 1.1

Viscosity: 400 (mPas)

Saponification value: 45-55 (mg KOH/g)

Acid value: ≤ 2.0(mg KOH/g)

HLB value: 14.9

Hydroxyl value: 81-96

Moisture content: 3.0 %

Solubility

It is soluble in water and ethanol. It is insoluble in mineral oil and vegetable oil.

Applications

- It is non-ionic surfactant widely used as emulsifying agent in the preparation of stable oil-in-water pharmaceutical emulsions.
- May also be used as solubilising agent for a variety of substance including essential oils and oil soluble vitamins.
- As wetting agent in the formulation of oral and parenteral suspensions.

Stability and Storage Conditions

Polysorbates are stable to electrolytes and weak acids and bases. The oleic acid esters are sensitive to oxidation.

Polysorbates are hygroscopic. It should be stored in a well closed container, protected from light, in a cool and dry place.

POLYETHYLENE GLYCOL 400[126]

Non-proprietary Names

BP: Macrogols

JP: macrogols 400

PhEur: Macrogols

USP-NF: Polyethylene Glycol

Synonym

Carbowax, carbowax sentry, Lipoxol, macrogola, PEG, Polyoxyethylene glycol, pluriol E

Chemical Name

α – Hydro – ω- hydroxypoly (oxy-1, 2-ethanediyl)

CAS Registry Number

25322-68-3

Empirical Formula

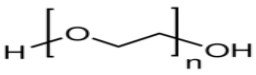
HOCH2 (CH2OCH2) mCH2OH

Where m = 8.7 (average number of oxyethylene groups) H (OCH2CH2) Noh

Molecular Weight

380-420

Structure



Functional Category

Ointment base, Plasticizer, Solvent, Suppository base, Tablet and capsule lubricant.

Description

It occurs as clear, colourless or slightly yellow colored viscous liquids.

Typical Properties

Specific gravity: 1.1254

Density: 1.11-1.14 g/cm3

Refractive index: 1.465

Viscosity: 90.0 (mPas)

Hydroxyl value: 264-300

Freezing point: 4-8 0C

Solubility

All grades of PEG are soluble in water. Liquid PEG is soluble in acetone, alcohols, benzene, glycerin and glycols.

Applications

- PEG's is widely used in a variety of pharmaceutical formulations including parenteral, topical, ophthalmic, oral and rectal preparations.
- Aqueous PEG solutions can be used either as suspending agents or to adjust the viscosity of other suspending vehicles.
- PEG can acts as an emulsion stabilizer.
- PEG can also be used to enhance the aqueous solubility or dissolution characteristics of poorly soluble compounds by making solid dispersions.

Stability and Storage Conditions

PEG is chemically stable in air and in solution. PEG does not support microbial growth and they do not become rancid.

PEG should be stored in well closed containers in a cool, dry place. Stainless steel, aluminium, glass or lined steel containers are preferred for the storage of liquid grades.

SPAN 80[127]

Non-proprietary Names

BP: Sorbitan monooleate 80

JP: Sorbitan monooleate 80

PhEur: Sorbitan monooleate 80

USP-NF: Sorbitan monooleate 80

Synonym

Sorbitan monooleate, Sorbitan, mono-(9Z)-9-octadecenoate, Arlacel 80, Span 80.

Chemical Name

Sorbitan mono-(9Z)-9-octadecenoate

CAS Registry Number

1338-43-8

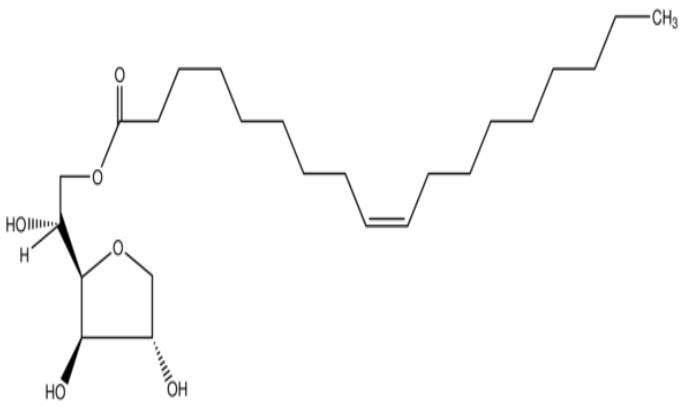
Empirical Formula

$C_{24}H_{44}O_6$

Molecular Weight

428.6

Structure



Functional Category

Dispersing agent, emulsifying agent, Non-ionic surfactant, solubilising agent, suspending agent and wetting agent

Description

Span has a characteristic odour and a warm, somewhat bitter taste. It is a yellow oily liquid.

Typical Properties

Specific gravity: 1.1

Viscosity: 970-1080 (mPas)

Saponification value: 145-160(mg KOH/g)

Acid value: ≤ 8.0(mg KOH/g)

HLB value: 4.3

Hydroxyl value: 193-209

Moisture content: 0.5 %

Solubility

It is soluble in water and ethanol. It is insoluble in mineral oil and vegetable oil.

Applications

- It is non-ionic surfactant widely used as emulsifying agent in the preparation of stable oil-in-water pharmaceutical emulsions.
- May also be used as solubilising agent for a variety of substance including essential oils and oil soluble vitamins.
- As wetting agent in the formulation of oral and parenteral suspensions.

Stability and Storage Conditions

Span is stable to electrolytes and weak acids and bases. The oleic acid esters are sensitive to oxidation.

Span is hygroscopic. It should be stored in a well closed container, protected from light, in a cool and dry place.

SPAN 60[127]

Non-proprietary Names

BP: Sorbitan monostearate 60

JP: Sorbitan monostearate 60

PhEur: Sorbitan monostearate 60

USP-NF: Sorbitan monostearate 60

Synonym Sorbitan stearate, Sorbitan monooctadecanoate, Sorbitan stearate (INN), Span 60 (TN)

Chemical Name

2-[(2R, 3S, 4R)-3, 4-dihydroxyoxolan-2-yl]-2-hydroxyethyl octadecanoate

CAS Registry Number

1338-41-6

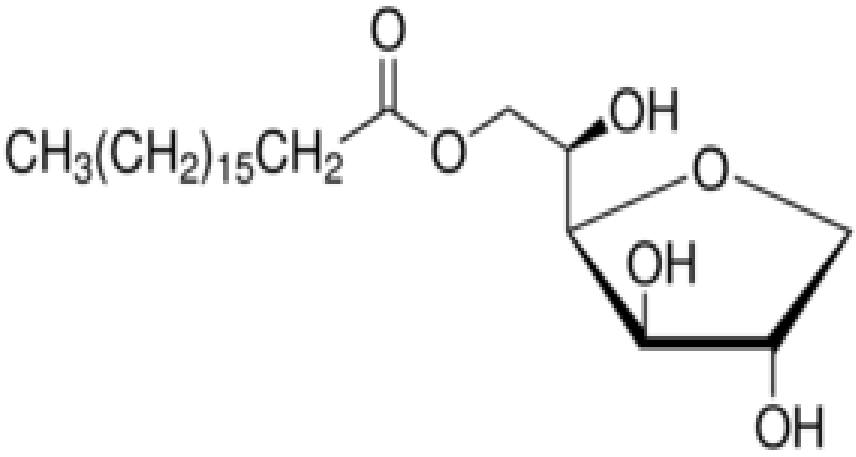
Empirical Formula

$C_{24}H_{46}O_6$

Molecular Weight

430.62

Structure



Functional Category

Dispersing agent, emulsifying agent, Non-ionic surfactant, solubilising agent, suspending agent and wetting agent

Description

Span has a characteristic odour and a warm, somewhat bitter taste. It is a yellow oily liquid.

Typical Properties

Saponification value: 147-157(mg KOH/g)

Acid value: 5-10(mg KOH/g)

HLB value: 4.7

Hydroxyl value: 235-260

Moisture content: 0.5 %

Solubility

It is soluble in water and ethanol. It is insoluble in mineral oil and vegetable oil.

Applications

- It is non-ionic surfactant widely used as emulsifying agent in the preparation of stable oil-in-water pharmaceutical emulsions.
- May also be used as solubilising agent for a variety of substance including essential oils and oil soluble vitamins.
- As wetting agent in the formulation of oral and parenteral suspensions.

Stability and Storage Conditions

Span is stable to electrolytes and weak acids and bases. The oleic acid esters are sensitive to oxidation.

Span is hygroscopic. It should be stored in a well closed container, protected from light, in a cool and dry place.

MATERIALS AND METHODS

8. MATERIALS AND METHODS

MATERIALS USED IN FORMULATION:

List of Materials Used

S.NO	NAME	CATEGORY	SOURCE
1	Ivermectin	API	Bafna pharmaceuticals, HP
2	Olive oil	Oil	Bought from market
3	Castor oil	Oil	Bought from market
4	Coconut oil	Oil	Bought from market
5	Rose oil	Oil	Bought from market
6	Tween 20	Surfactant	Madras pharma, Chennai
7	Tween 60	Surfactant	Madras pharma, Chennai
8	Tween 80	Surfactant	Madras pharma, Chennai
9	PEG-400	Co-surfactant	Madras pharma, Chennai
10	Span 60	Co-surfactant	Madras pharma, Chennai
11	Span 80	Co-surfactant	Madras pharma, Chennai

Table 2 List of materials used

EQUIPMENTS / INSTRUMENTS USED IN FORMULATION

List of Equipments / Instruments Used

S.NO	Equipments / Instruments	Equipments / Instruments
1	Electronic weighing balance	M.C.Dalal, Chennai
2	UV-Visible spectrophotometer	Jasco V-630 / Shimadzu 1800
3	FT-IR	Shimadzu,Japan
4	Vortex mixer	Remi CM 10, Mumbai
5	Orbital shaker	Scigrnics biotech, Orbitek
6	Magnetic stirrer	Remi, Mumbai
7	Cooling centrifuge	Remi, Mumbai
8	Particle size analyzer	Malvern instruments, UK
9	Zeta potential analyzer	Malvern instruments, UK
10	pH meter	M.C.Dalal, Chennai
11	Hot air oven	M.C.Dalal, Chennai
12	Dissolution apparatus	Thermionic, Campbell Electronics
13	0.45 µ filter	Merck Millipore
15	Refractometer	Atago Rx-7000i, Japan

Table 3. List of Equipments / Instruments Used

METHODOLOGY:

DRUG EXCIPIENTS COMPATIBILITY STUDY[25, 51, 71]:

The drug and excipients selected for the formulation are evaluated for compatibility study.

Chemical Compatibility Study[111, 112]:

For this study Infrared spectroscopic method was used. The spectroscopic studies were carried out to find the interaction between pure drug, Oil, Surfactants, Co-Surfactants, Adsorbent and its physical mixture by KBr pellet technique and Nujol mull technique using FT-IR spectrophotometer. The IR spectrum of the each excipient was compared with the IR spectrum of pure drug. The spectrum was recorded in the range of 4000 – 500 cm-1.

CALIBRATION CURVE OF IVERMECTIN

Selection of solvent:

Ivermectin was freely soluble in ethanol which was used as the solvent to solubilise the standard drug and the sample as well.

Determination of λmax:

A stock solution was prepared by dissolving an approximate quantity of (100 mg) of pure drug Ivermectin in ethanol and made up to 100 ml with to obtain (1000µg/ml).The stock solution of ivermectin was diluted with the solvent to get a concentration of 10 µg/mL. This solution was scanned in the UV region from 400 to 200 nm.

Preparation of Calibration Curve:

The standard stock solution of ivermectin was prepared by dissolving 10 mg of the drug in 10 mL of the solvent to obtain a concentration of 1 mg/mL. The above solution was further diluted to get concentrations in the range of 5–15 µg/mL. The absorbance of the solutions was measured by using UV-Visible spectrophotometer at 245 nm against blank. Calibration curve was plotted by using concentration on X-axis and absorbance on Y-axis.

INTRINSIC DISSOLUTION STUDY OF IVERMECTIN[32]:

An equivalent of single dose of IVERMECTIN (i.e. 6 mg) was taken in the hard gelatin capsule (size 5). The dissolution study was carried out in the USP type I apparatus (Basket type). 0.1 N HCl was used as dissolution medium and basket is rotated at 75 rpm the medium was maintained at 37 ± 0.50C. The 10 ml of samples were collected at 15 minutes interval. Then the concentrations of samples were obtained using UV –Visible spectrophotometer at 245 nm against blank.

SOLUBILITY OF IVERMECTIN[116]:

The screening of different vehicles is a prerequisite for the formulation of SMEDDS. The solubility of Ivermectin in various oils, surfactants and co-surfactants was determined by using shake flask method. In 2 ml of each of selected oil, surfactant and co-surfactant an excess amount (200 mg) of drug was added in sealed vials. The components were mixed by using vortex mixer for 10 minutes and then kept in orbital shaker for 72 hrs to attain equilibrium. The orbital shaker was set for 100 rpm at room temperature. After equilibrium samples were centrifuged for 15 minutes at 5000 rpm the resulted supernatant was diluted suitably with methanol. The concentration of Ivermectin was quantified using UV-Visible spectrophotometer at 245 nm.

OPTIMIZATION OF IVERMECTIN SMEDDS USING D- OPTIMAL DESIGN[61, 62]**:**

A D-optimal design was developed to statistically optimize the formulation factors and evaluate the main effects, interaction effects and quadratic effects on the independent factors. 3 factors were used to explore quadratic response surfaces and constructing second order polynomial models with design expert (version 8.0.6, Stat Ease Inc.)., and a matrix comprising 3 factors, 3 level is selected for the optimization study. The experimental design consists of a set of points lying at the midpoint of each edge and the replicated centre point of a multidimensional cube. Independent and dependent variables are listed in Table. The polynomial equation generated by this experimental design (using sigma plot 11) is as follows:

$$Yi= \beta0 + \beta1X1 + \beta\ 2X2 + \beta3X3 + \beta12X1X2 + \beta13X1X3 + \beta23X2X3 + \beta11X12+ \beta22X22+ \beta33X32$$

Where Yi is the dependent variable, β0 is the intercept, β1 to β33 are the regression co-efficient, and x1, x2 and x3 are the independent variables selected from preliminary experiments.

Optimization validation and data analysis:

Statistical validation of the poly nominal equation and ANOVA was calculated using Design Expert software. The resultant experimental values of the responses were quantitatively compared with the predicted values to calculate the prediction error.

D- optimal mixture design was used for the optimization of Ivermectin loaded SMEDDS formulation. The amount of oil, surfactant and co-Surfactant were the three factors (Independent variables) studied. The responses (Dependent variables) studied were Self emulsification time, Globule size and % transmittance.

Summary of Experimental Design

INDEPENDENT VARIABLES	**UNITS**	**Level**		
		Low(-1)	**Medium(0)**	**High(+1)**
X1= Oil	% W/V	10	15	20
X2= Surfactant	% W/V	20	35	50
X3= Co-surfactant	% W/V	20	35	50
Dependent variables	**Units**	**Constraints**		
Self emulsification time	Seconds	Minimize		
Globule size	manometer	Minimize		
% Transmittance	%	Maximize		

Table 4. Summary of Experimental Design

CONSTRUCTION OF TERNARY PHASE DIAGRAMS[117, 118]:

Ternary phase diagram is useful to identify the best emulsification region of oil, surfactant and co-surfactant combinations. Ternary phase diagram of surfactant, co-surfactant and oil are plotted; each of them, representing an apex of the triangle 9. The methods used to plot Ternary phase diagrams are namely dilution method and water Titration method.

a) Dilution Method

A ternary mixture with varying compositions of surfactant, co-surfactant and oil is prepared. The surfactant concentration is varied from 20 to 50% (w/w), oil concentration is varied from 10 to 20% and co-surfactant concentration is varied from 20 to 50% (w/w). For any mixture, the total of surfactant, co-surfactant and oil concentrations is always added to 100%.

Further, the co-surfactant was increased by 5% for each composition, oily phase concentration kept constant and the surfactant concentration adjusted to make a total of 100%. Forty-two such mixtures with varying surfactant, co-surfactant and oil concentrations are prepared. Compositions are evaluated for micro emulsion formation by diluting appropriate amount of 42 mixtures with appropriate double distilled water. Globule size of the resulting dispersions is determined by using spectroscopy technique. Dispersions, having globule size 100 nm or below are considered desirable. The area of micro emulsion formation in Ternary phase diagram is identified for the respective system in which micro emulsions with desire globule size are obtained.

Water Titration Method

The pseudo-ternary phase diagrams are also constructed by titration of homogenous liquid mixtures of oil, surfactant and co-surfactant with water at room temperature. Oil phase, Surfactant and the co-surfactant, at Km values 1.5 and 1 (surfactant: co-surfactant ratio), oily mixtures of oil, surfactant and co-surfactant are prepared varied from 9:1 to 1:9 and weighed in the same screw-cap glass tubes and are vortexed. Each mixture was then slowly titrated with aliquots of distilled water and stirred at room temperature to attain equilibrium. The mixture visually examined for transparency. After equilibrium was reached, the mixtures are further titrated with aliquots of distilled water until they showed the turbidity. Clear and isotropic samples are deemed to be within the micro emulsion region. No attempts were made to completely identify the other regions of the phase diagrams. Based on the results, appropriate percentage of oil, surfactant and co-surfactant was selected, correlated in the phase diagram and were used for preparation of SMEDDS.

FORMULATION DEVELOPMENT[62]:

Formulation of L-SMEDDS

A series of SMEDDS formulations were prepared using selected oil, surfactant and co-surfactant based on solubility study reports. All the formulations were prepared using 6 mg of Ivermectin. The resultant mixture was vortexed and heated at 40◦C until a clear solution was obtained. The SMEDDS formulations were stored in sealed vials at room temperature for further use.

CHARACTERIZATION OF L- SMEDDS FORMULATIONS[117, 118]:

Determination of Self-Emulsification Time

The efficiency of self-emulsification was estimated by using USP type II (paddle) at 100 rpm. 0.1 N HCl was used as emulsification medium and maintained at 37±0.50C. 1 ml of L-SMEDDS was poured in 100 ml of the medium drop wise and paddle was rotated at 100 rpm. The self-emulsification time was noted for each formulation.

Determination of % Transmittance (% T):

The L-SMEDDS formulations were diluted to 100 folds with distilled water. The % transmittance of the prepared emulsion was measured at 650 nm using UV-Visible spectrophotometer. Distilled water was used as blank.

Determination of Refractive Index (RI)

The clarity of prepared L-SMEDDS could be used to access the refractive index. The refractive index was measured using Atago Rx-7000i and distilled water was used as standard (RI = 1.3332 0 at 25◦C)

Determination of Viscosity[117-119]:

a) **Before dilution**

The viscosity of the prepared micro emulsion formulations were determined as such without dilution by Brookfield DV III ultra V6.0 RV cone and plate rheometer (Brookfield Engineering Laboratories, Inc, Middleboro, MA) using spindle # CPE40 at 25°C ±0.5°C. The software used for the viscosity calculations was Rheocalc V2.6

Table 5.Parameters specification for Viscosity determination

Parameters	Optimized specification
Sample (g)	0.5
Speed (rpm)	6
Data Interval (min)	10
Loop Start	CP-41
Wait time (min)	5
Temperature (°C)	25±0.5
Share rate (1/sec)	7.5(N)

b) After dilution with water:

The viscosity of diluted SMEDDS was determined. The viscosity of Diluted SMEDDS shows the type of emulsion formed. Low viscosity indicates the formation of w/o emulsion and high viscosity indicates o/w emulsion**.**

Determination of pH[119]:

pH values of SMEDDS were determined using pH meter.

Drug Content Study of L-SMEDDS[119, 120]:

An amount of L-SMEDDS equivalent to 6 mg was carefully weighed and transferred into a 100 ml standard flask and diluted to 100 ml with 0.1 N HCl. The resulting solution was filtered through 0.45 µ filter. 1 ml of the clear solution was taken and diluted to 100 ml with 0.1 N HCl. The concentration of the resulting solution was measured at 245 nm by UV-Visible spectrophotometer against blank. The limit for drug content is not less than 90.0% and not more than 110.0%.

Dispersibility tests[116]:

The efficiency of self-emulsification of oral micro emulsion was assessed using a standard USP XXII dissolution apparatus 2. One ml of each formulation was added to 500 mL of distilled water, 0.1N HCl and Phosphate buffer pH 6.8 respectively at 37 ± 0.5o C. A standard stainless steel dissolution paddle rotating at 50 rpm provided gentle agitation. The in

vitro performance of the formulations was visually assessed using the following grading system. Those formulations that passed the thermodynamic stability and also dispersibility test in Grade A were taken for further studies. Further from each Smix Group one formulation is selected, having the least Smix concentration irrespective of Smix ratio used, but passing dispersibility test in Grade A in distilled water, Phosphate buffer pH 6.8 as well as in 0.1N HCl**.**

Table 6. Emulsification Study

S.NO	Grade	Appearance
1	A	Rapidly forming (within 1 min) emulsion, white a clear or bluish appearance
2	B	Rapidly forming (within 1 min) slightly less clear emulsion, with a bluish white appearance
3	C	Fine milky emulsion that formed within 2 min
4	D	Dull, greyish white emulsion having slightly oily appearance that is slow to emulsify (longer than 2 min)
5	E	Formulation exhibiting either poor or minimal emulsification with large oil droplets on the surface

Globule Size and Zeta potential analysis[121]:

The mean globule sizes (z-average), Zeta potential (ζ) as well as the Polydispersity index (PDI) of emulsions formed from stable SMEDDS formulations were determined by using Malvern zetasizer (version 7.11 Malvern instruments UK). Before the analysis each formulation was diluted to a suitable concentration with distilled water (100 times with distilled water) i.e. 1 ml to 100 ml. Size analysis was performed at 25ºC with an angle of detection of 90º. The principle involved is due to Brownian motion of droplets as a function of time which is determined due to fluctuation in light scattering and it determines by photon correlation spectroscopy.

In-vitro drug release performance[116-120]:

The study was performed by using dialysis bag method

Dialysis membrane specification

The dialysis membrane used in the study was Cellulose membrane (Sigma, USA). Tubing as such without treatment is stored at room temperature. Its capacity was 60 mL/feet; average flat width was 2.5 mm, diameter and 16 mm.

Treatment of dialysis bag

a) Soak the Dialysis bag in Glycerin for 15 minutes

b) Remove the glycerin by washing in running water for 3-4 minutes.

c) Remove sulphur compounds by treating it with 0.3% w/v sodium sulphide solution in water at 80º C for 1 minute.

d) Wash with hot water at 60º C for 2 minutes.

e) Acidify the procured dialysis bag with 0.2% v/v H2SO4 in distilled water.

f) Rinse it with hot water to remove acid.

g) Store the dialysis bag in the dissolution medium in refrigerator in which the dissolution experiments are carried out, so that the pores remain open.

In-vitro drug release study:

In vitro release test was performed in 900 ml of 0.1 N HCl, which (Dissolution apparatus I.P. 2, at 100 rpm and 37 ± 0.5 0 C). Single dose containing 6 mg (Containing 6 mg of Ivermectin) was placed in treated dialysis bag. Samples are withdrawn at periodic intervals and same amount of buffer is replaced.

***In Vitro* Drug Release Study of Marketed Formulation[69]**

For the *in vitro* drug release study, USP type 2 apparatus (paddle type) was used. The marketed formulation was taken and placed into the medium. 900 ml of 0.1N HCl maintained at 37±0.5^{0}C and stirred at 75 rpm was used as dissolution medium. 10 ml of samples were withdrawn at predetermined time intervals (i.e. 5 min) and replaced with equivalent amount of fresh medium. Samples were filtered through 0.45 µm filter. The concentrations of samples were determined by using UV-Visible spectrophotometer at 245 nm against blank.

Comparison of *In Vitro* Drug Release of API, L-SMEDDS and Marketed Formulation of Ivermectin.

The *in vitro* drug release of API, L-SNEDDS formulation and marketed tablets were done using USP dissolution apparatus and the values were noted.

***IN VITRO* RELEASE KINETICS OF OPTIMIZED FORMULATION[69]**

To study the *in vitro* release kinetics of the optimized formulation, data obtained from dissolution study were plotted in various kinetics models.

1. Zero Order Equation

   The zero order release can be obtained by plotting cumulative percentage drug released vs. Time in hours

   $C=K0t$

   Where, K0 = Zero order constant

   t= time in hours.

2. First Order Reaction

   The graph was plotted as % cumulative drug release vs. Time in hours

   $\text{Log } C = \text{Log } C0 - Kt / 2.303$

   Where, C0 = initial concentration of drug,

   K = First order

   t = time in hours.

3. Hixson and Crowell Erosion equation

   To evaluate the drug release with changes in the surface area and thediameter of the particles, the data were plotted using the Hixson and Crowell rate equation. The graph was plotted by cube root of % drug remaining vs.Time in hours.

$$Q_0^{1/3} - Q_t^{1/3} = K_{HC}t$$

Where, Q_0 = Initial amount of drug

Q_t = Amount of drug released in time t,

K_{HC} = Rate constant for Hixson Crowell equation

4. Higuchi Kinetics

The graph was plotted with % cumulative drug release vs. Square root of time

Q = Kt1/2

Where, K = constant reflecting design variable system (differential rate constant) t = time in hours.

The drug release rate is inversely proportional to the square root of time.

5. Korsmeyer- Peppas Equation

To evaluate the mechanism of drug release, t was further plotted in Koresmeyer – Peppas equation as log cumulative % of drug released vs. log time

$Mt / M\alpha = Ktn$

Where, $Mt / M\alpha$ = Fraction of rug released at time t

t = Release time

K = kinetics constant (instructing structural and geometric characteristic of the formulation)

N= Diffusion exponent indicative of the mechanism of drug release.

Table 7. Diffusion Exponent and Solute Release Mechanism for Cylindrical Shape Diffusion

Diffusion Coefficient	Overall solute diffusion mechanism
0.45	Fickian diffusion
0.45< n <0.89	Anomalous (non-Fickian) diffusion
0.89	Case II transport
N >0.89	Super case II transport

ACCELERATED STABILITY STUDIES

The design of stability studies to support submission of NDAs and MAAs are described in the ICH guidance Q1A (R2) stability testing of new drug substance and products(3).

The stability test for SNEDDS formulations are performed as per the ICH guidance which are described in Table.17.

Table 8. ICH guidance description for stability study of pharmaceutical Formulations.

Study	Storage condition	Minimum periods
Long Term	$25^0C \pm 2^0C$ / 60% RH ± 5% RH or $30^0C \pm 2^0C$ / 65% RH ± 5% RH	12 Months
Intermediate	$30^0C \pm 2^0C$ /65% RH ± 5% RH	6 Months
Accelerated	$40^0C \pm 2^0C$ / 75% RH ± 5% RH	6 Months

RESULTS AND DISCUSSION

9. RESULTS AND DISCUSSION

DRUG – EXCIPIENTS COMPATIBILITY STUDY

Chemical Compatibility Study

The possible interactions between the drug and excipients used in the formulation were studied by FT-IR spectroscopy. The results are given in Fig 6 to 9 and Table 9 to 12.

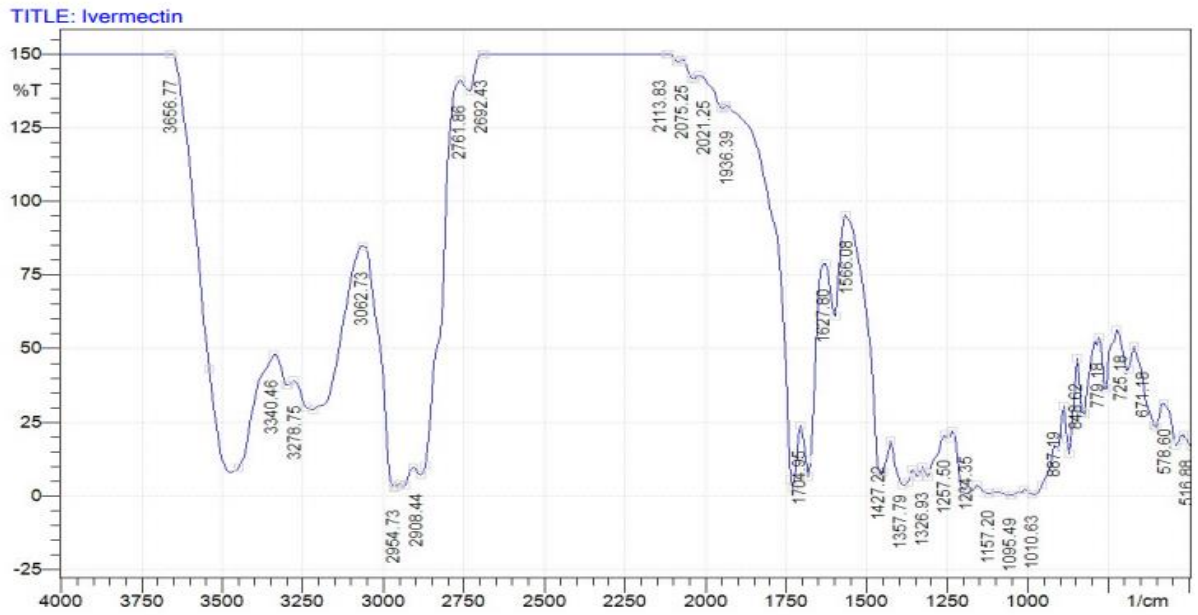
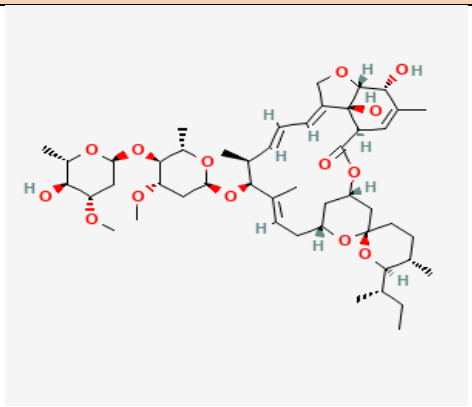


Fig 6. FTIR Spectrum of Ivermectin

Table 9. FTIR Interpretation of Ivermectin

STRUCTURE	FUNCTIONAL GROUP	WAVE NUMBER (observed)
	O-H stretching	3340 cm^{-1}
	C=O stretching	1704 cm^{-1}
	C-O-C stretching	1157 cm^{-1}
	C=C stretching	1627 cm^{-1}
	Alkane SP^3 C-H	2908 cm^{-1}
	Alkane SP^2 C-H	3062 cm^{-1}

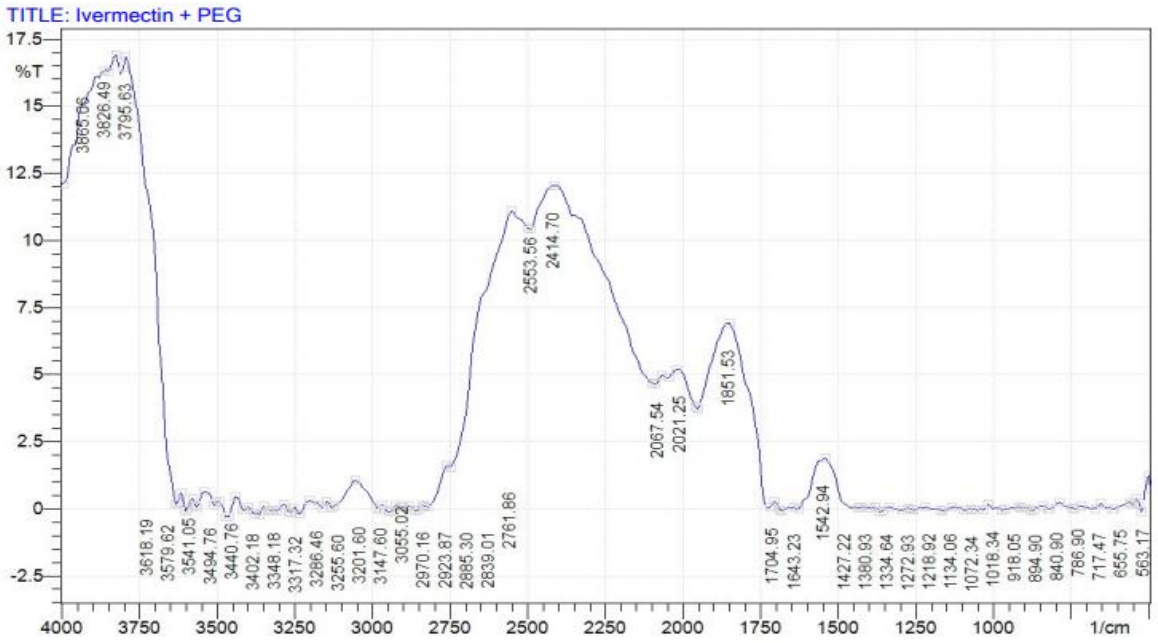


Fig 7. FTIR Spectrum of Ivermectin and PEG 400.

Table 10. FTIR Interpretation of Ivermectin and PEG 400.

FUNCTIONAL GROUP	WAVE NUMBER (observed)
O-H stretching	3340 cm^{-1}
C=O stretching	1704 cm^{-1}
C-O-C stretching	1134 cm^{-1}
C=C stretching	1643 cm^{-1}
Alkane SP^3 C-H	2923 cm^{-1}
Alkene SP^2 C-H	3055 cm^{-1}

The peak observed in the FT-IR spectrum of Ivermectin and Poly Ethylene Glycol showed no shift and no disappearance of the characteristic peaks of drug. This suggests that there is no interaction between the drug and Poly Ethylene Glycol.

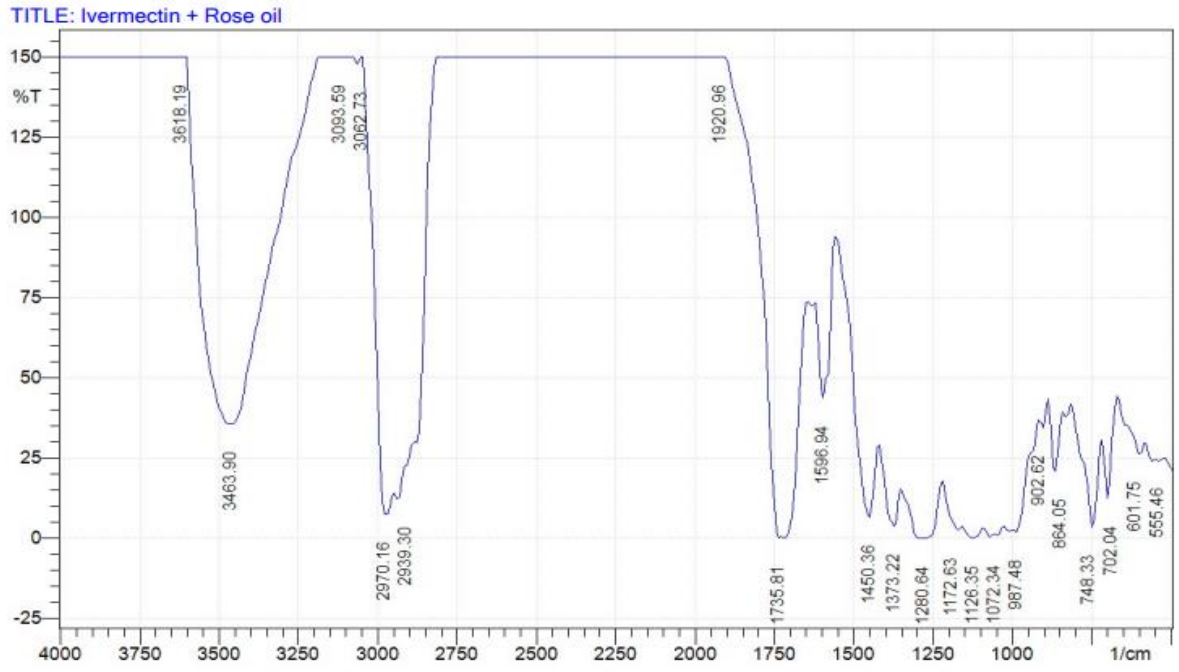


Fig 8. FTIR Spectrum of Ivermectin and Rose oil.

Table 11. FTIR Interpretation of Ivermectin and Rose oil.

FUNCTIONAL GROUP	WAVE NUMBER (observed)
O-H stretching	3463 cm^{-1}
C=O stretching	1735 cm^{-1}
C-O-C stretching	1172 cm^{-1}
C=C stretching	1596 cm^{-1}
Alkane SP^3 C-H	2939 cm^{-1}
Alkene SP^2 C-H	3062 cm^{-1}

The peak observed in the FT-IR spectrum of Ivermectin and Rose oil showed no shift and no disappearance of the characteristic peaks of drug. This suggests that there is no interaction between the drug and Rose oil.

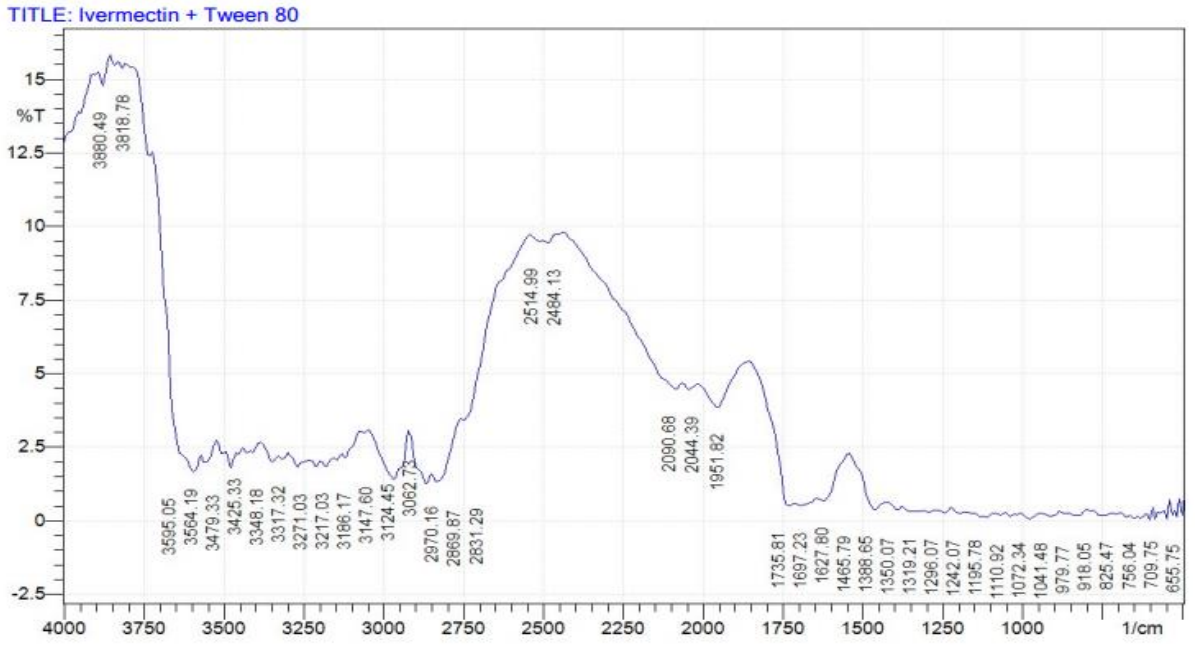


Fig 9. FTIR Spectrum of Ivermectin and Tween 80.

Table 12. FTIR Interpretation of Ivermectin and Tween 80.

FUNCTIONAL GROUP	WAVE NUMBER (observed)
O-H stretching	3348 cm^{-1}
C=O stretching	1735 cm^{-1}
C-O-C stretching	1195 cm^{-1}
C=C stretching	1627 cm^{-1}
Alkane SP^3 C-H	2970 cm^{-1}
Alkene SP^2 C-H	3062 cm^{-1}

The peak observed in the FT-IR spectrum of Ivermectin and Tween 80 showed no shift and no disappearance of the characteristic peaks of drug. This suggests that there is no interaction between the drug and Tween 80.

CALIBRATION CURVE OF IVERMECTIN

The UV-Visible spectroscopic method was used to analyze the calibration curve of Ivermectin. The absorbance of the drug in 0.1N HCL was measured at a wavelength of 245 nm against blank. The results are given in table 13 and Fig 10.

Table 13. Data for Calibration Curve of Ivermectin

S.No.	Concentration (µg/ml)	Absorbance
1	2	0.1523±0.0012
2	4	0.3410±0.0086
3	6	0.5101±0.0014
4	8	0.6751±0.0098
5	10	0.8480±0.0155
R^2		0.9997

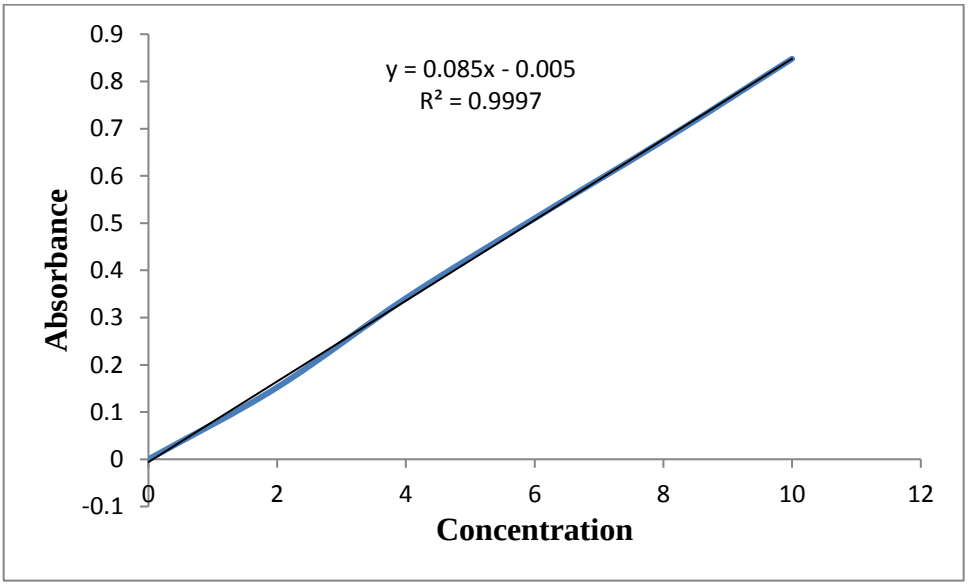


Fig 10. Calibration curve of Ivermectin

It is found that the solution of Ivermectin in 0.1 N HCl (pH-1.2) obeys linearity in the concentration range of up to 15 µg/ml.

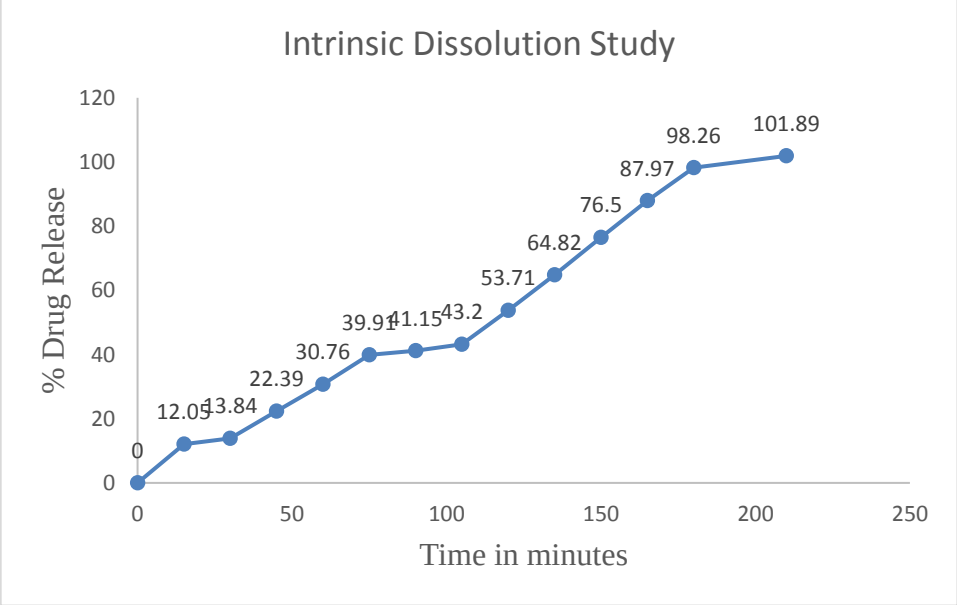
INTRINSIC DISSOLUTION STUDY OF IVERMECTIN

The intrinsic dissolution study was performed by using USP type II dissolution apparatus (Basket). Results of the study are presented in the Table 14 and Fig 11.

Table 14. Intrinsic dissolution study of Ivermectin

S.No	Time (min)	% Drug release
1	15	12.05
2	30	13.84
3	45	22.39
4	60	30.76
5	75	39.91
6	90	41.15
7	105	43.20
8	120	53.71
9	135	64.82
10	150	76.50
11	165	87.97
12	180	98.26
13	210	101.89

Fig.11 Intrinsic dissolution study of Ivermectin



SOLUBILITY OF IVERMECTIN

The high solubility of drug in the oil phase is the crucial parameter in designing stable SMEDDS formulations. The drug should possess good solubility in solvent, so precipitation during the shelf life of the formulation and after dilution in water phase can be avoided. The solubility of Ivermectin in selected oils, surfactants and co-surfactants is presented in Table 13.

Table 15. Solubility Study of Ivermectin

S.NO	SOLVENTS	SOLUBILITY (mcg/ml)
OILS		
1	Castor oil	729.36
2	Coconut oil	849.80
3	Olive oil	740.20
4	Rose oil	912.00
SURFACTANT		
5	Tween 80	984.25
6	Tween 60	708.94
7	Tween 20	840.34
CO-SURFACTANT		
8	Span 80	548.12
9	Span 60	678.22
10	PEG-400	828.22

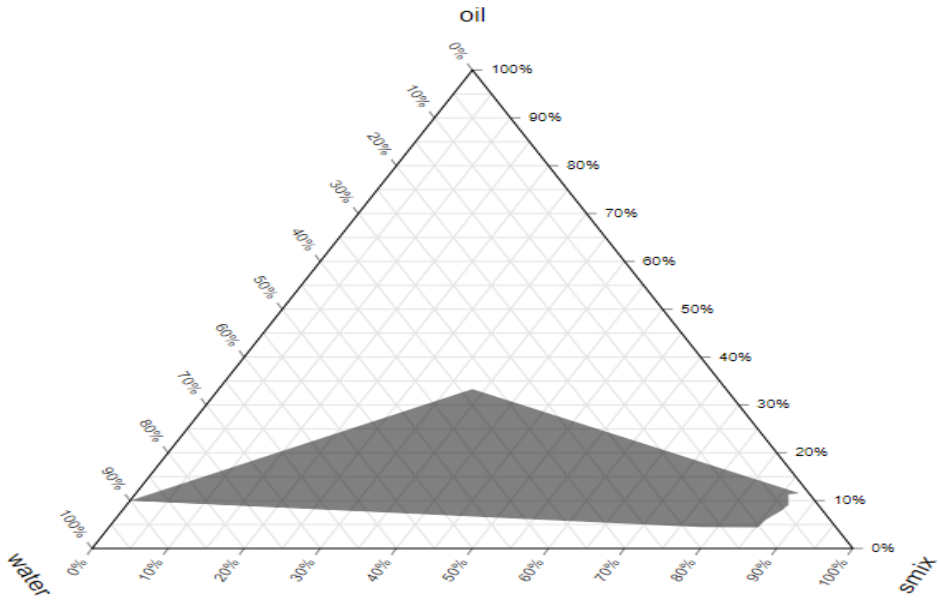
The solubility of Ivermectin was observed in oils, the solubility in Castor oil was 729.36 mcg/ml, the solubility in Coconut oil was 849.80 mcg/ml, in Olive oil 740.20 mcg/ml and the solubility in rose oil was 912.00 mcg/ml. The solubility of Ivermectin was observed in surfactants, the solubility in Tween 80 was 984.25 mcg/ml, the solubility in Tween 60 was 708.94 mcg/ml and the solubility in Tween 20 is 840.34 mcg/ml. The solubility of Ivermectin was observed in co-

surfactants, the solubility in Poly ethylene glycol is 828.22 mcg/ml, the solubility in Span 60 is 678.22 mcg/ml and the solubility in Span 80 is 548.12 mcg/ml. The maximum solubility of Ivermectin was observed in Rose oil (912.00 mcg/ml), Tween 80 (984.25 mcg/ml) and PEG 400 (828.22 mcg/ml). The components provided the best solubility of Ivermectin were further used to develop the L-SMEDDS formulations. Surfactants can cause gastrointestinal irritation, so their selection is very important factor in SMEDDS design. The non-ionic surfactants are less toxic than ionic ones and they are characterized by lower micelle concentration values. Another important criterion is surfactant with proper hydrophilic-lipophilic balance (HLB) value. Generally surfactants with HLB values of 12 to 15 recommended for the preparation of SMEDDS. The HLB value of selected surfactant is 14 to 16.

CONSTRUCTION OF TERNARY PHASE DIAGRAM

The ternary phase diagram was constructed using ternary plot generator and found out the region of w/o type micro emulsion found out by water titration method. Various ratio of Oil, Smix were titrated with water and found the micro emulsion region was found

Fig. 12. Ternary phase diagram



From the ternary phase diagram micro emulsion region was identified by using water titration method. From the observed micro emulsion region independent variables for D- Optimal design are identified.

D- OPTIMAL DESIGN From the ternary diagram, the ranges for each component were selected as follows: 20%-50% surfactant, 20%-50%, and co surfactant,10%-20% oil, response factors were used to assess the quality of the SMEDDS formulation, including self emulsification time (Y1), globule size (Y2) and % transmittance (Y3) At three-factor, D-Optimal requires 16 experimental runs with four central points to determine the experimental error and the precision of the design. A total of 16 experimental runs were generated and evaluated using Design-Expert software (V. 8.0.6; Stat-Ease Inc., Minneapolis, Minnesota).

Std	Run	Factor 1Oil	Factor 2 Surfactant	Factor 3 Co-surfactant	Response 1 Self emulsificationtime	Response 2 Globule size	Response 3 % Transmittance
1	1	19.89211	44.94211	35.16578	42.09	185.65	97.89
2	2	14.12332	50	35.87668	25.36	170.45	98.25
3	3	14.35518	45.51713	40.12769	27.89	173.45	98.9
4	4	10	44.9	45.1	20.15	161.78	99.25
5	5	14.17891	35.82109	50	26.35	170.87	98.54
6	6	20	50	30	52.48	189.45	96.96
7	7	18.49734	40.65644	40.84622	34.58	180.98	97.79
8	8	20	50	30	49.75	189.26	96.53
9	9	20	30	50	50.19	191.45	96.25
10	10	10	40.16848	49.83152	20.12	155.26	99.1
11	11	18.49734	40.65644	40.84622	31.25	178.65	97.89
12	12	14.36041	40.13959	45.5	25.19	167.58	98.78
13	13	20	30	50	61.23	191.12	96.56
14	14	14.17891	35.82109	50	26.69	169.87	98.56
15	15	14.12332	50	35.87668	27.45	159.32	98.67
16	16	20	35.00462	44.99538	54.76	192.58	97.23

Table 16. Actual summary of D-Optimal design for Ivermectin SMEDDS

Table 17. Design Summary

Factor	Name	Units	Low Actual	High Actual	Low Coded	High Coded	Mean	Std. Dev.
A	Oil	% W/V	10	20	-1	1	15	3.92232
B	Surfactant	% W/V	20	50	-1	1	35	11.767
C	Co-Surfactant	% W/V	20	50	-1	1	35	10.9464

Response 1: Self Emulsification Time **Transform: None**

Table 18. Sequential Model Sum of Squares [Type I]

Sequential Model Sum of Squares [Type I]						
Source	**Sum of Squares**	**df**	**Mean Square**	**F Value**	**p-value Prob > F**	
Mean vs Total	20702.17	1	20702.17			
Linear vs Mean	2221.856	2	1110.928	26.62465	< 0.0001	
Quadratic vs Linear	367.392	3	122.464	6.996335	0.0081	Suggested
Sp Cubic vs Quadratic	3.7772	1	3.7772	0.198495	0.6665	
Cubic vs Sp Cubic	85.57336	3	28.52445	1.997286	0.2159	
Quadratic vs Cubic	13.23609	1	13.23609	0.913419	0.3831	Aliased
Residual	72.45355	5	14.49071			
Sp Quadratic vs Quadratic	23.16521	3	7.721737	0.355899	0.7868	
Quadratic vs Sp Quadratic	79.42144	2	39.71072	2.740426	0.1572	Aliased
Residual	72.45355	5	14.49071			
Total	23466.46	16	1466.654			

"Sequential Model Sum of Squares [Type I]": Select the highest order polynomial where the additional terms are significant and the model is not aliased.

Table 19. Model Summary Statistics

Source	Std. Dev.	R-Squared	Adjusted R-Squared	Predicted R-Squared	PRESS	
Linear	6.459532	0.803772	0.773583	0.698137	834.4361	
Quadratic	4.183781	0.936678	0.905017	0.844575	429.639	Suggested
Special Cubic	4.36225	0.938044	0.896741	0.821149	494.3958	
Cubic	3.779101	0.969001	0.922503	-29.868	85328.04	
Special Quadratic	4.657942	0.945058	0.882268	0.328078	1857.387	
Quadratic	3.806667	0.973789	0.921368	+	Aliased	

+ Case(s) with leverage of 1.0000: PRESS statistic not defined.

"Model Summary Statistics": Focus on the model maximizing the "Adjusted R-Squared"and the "Predicted R-Squared".

ANOVA for Response Surface Quadratic Model

Table 20. Analysis of variance table [Partial sum of squares - Type III]

Source	Sum of Squares	Df	Mean Square	F Value	p-value Prob > F	Comments
Model	1864.163752	9	207.129306	29.050287	0.0092	significant
A-Oil	1347.8432	1	1347.8432	189.03762	0.0008	
B-Surfactant	0.538881059	1	0.53888106	0.0755791	0.8012	
C-Co-Surfactant	0.180780889	1	0.18078089	0.0253549	0.8836	
AB	0.893025	1	0.893025	0.1252485	0.7468	
AC	1.428025	1	1.428025	0.2002833	0.6848	
BC	200.7814433	1	200.781443	28.159987	0.0131	
A^2	64.0388206	1	64.0388206	8.9815688	0.0578	
B^2	7.213797418	1	7.21379742	1.0117491	0.3886	
C^2	188.3679858	1	188.367986	26.418975	0.0143	
Residual	21.39007845	3	7.13002615			
Cor Total	1885.553831	12				

The Model F-value of 29.58 implies the model is significant. There is only a 0.01% chance that a "Model F-Value" this large could occur due to noise.

Values of "Prob > F" less than 0.0500 indicate model terms are significant. In this case Linear Mixture Components, AB, AC, BC are significant model terms. Values greater than 0.1000 indicate the model terms are not significant. If there are many insignificant model terms (not counting those required to support hierarchy), model reduction may improve your model.

The "Lack of Fit F-value" of 1.42 implies the Lack of Fit is not significant relative to the pure error. There is a 35.60% chance that a "Lack of Fit F-value" this large could occur due to noise. Non-significant lack of fit is good -- we want the model to fit.

Std. Dev.	4.183781	R-Squared	0.936678
Mean	35.97063	Adj R-Squared	0.905017
C.V. %	11.6311	Pred R-Squared	0.844575
PRESS	429.639	Adeq Precision	14.25097

The "Pred R-Squared" of 0.8446 is in reasonable agreement with the "Adj R-Squared" of 0.9050.

"Adeq Precision" measures the signal to noise ratio. A ratio greater than 4 is desirable. Your

Ratio of 14.251 indicates an adequate signal. This model can be used to navigate the design space.

Component	Coefficient Estimate	df	Standard Error	95% CI Low	95% CI High	VIF
A-oil	61.44559	1	25.46653	4.702637	118.1885	37.51774
B-surf	56.87592	1	2.798517	50.64043	63.1114	2.088484
C-co surf	50.05484	1	2.79387	43.82971	56.27997	1.954737
AB	-152.77	1	46.07139	-255.423	-50.1163	22.75081
AC	-125.044	1	41.12555	-216.677	-33.4105	12.26631
BC	-37.2802	1	12.86931	-65.9548	-8.6056	1.800488

Final Equation in Terms of U_Pseudo Components:

S-M time = +61.45 * A+56.88 * B+50.05 * C -152.7 * A * B -125.0 * A * C-37.28 * B * C

Final Equation in Terms of Real Components:

S-M time =+2566.67990 * oil+346.70865* surf+242.18522* co surf-3819.24289* oil * surf

-3126.09890* oil * co surf-932.00537* surf * co surf

Final Equation in Terms of Actual Components:

S-M time =+25.66680* oil+3.46709 * surf+2.42185 * co surf-0.38192 * oil * surf-0.31261 * oil * co surf-0.093201 * surf * co surf

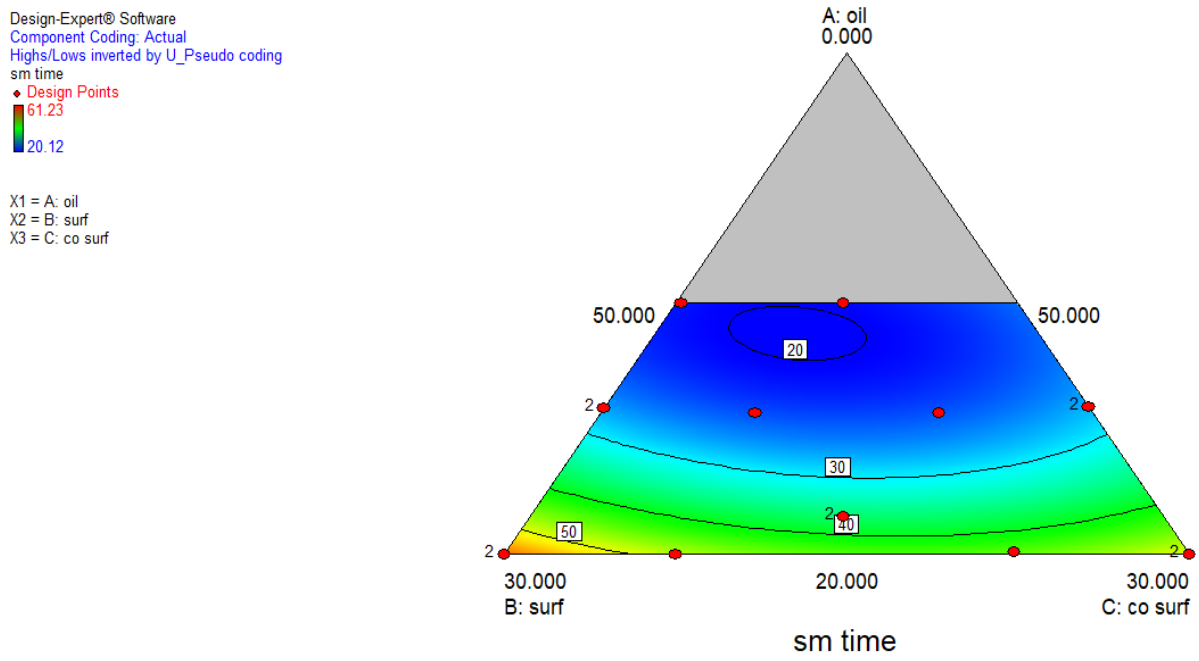


Fig 13. Contour Plot for Response 1(Self emulsification Time)

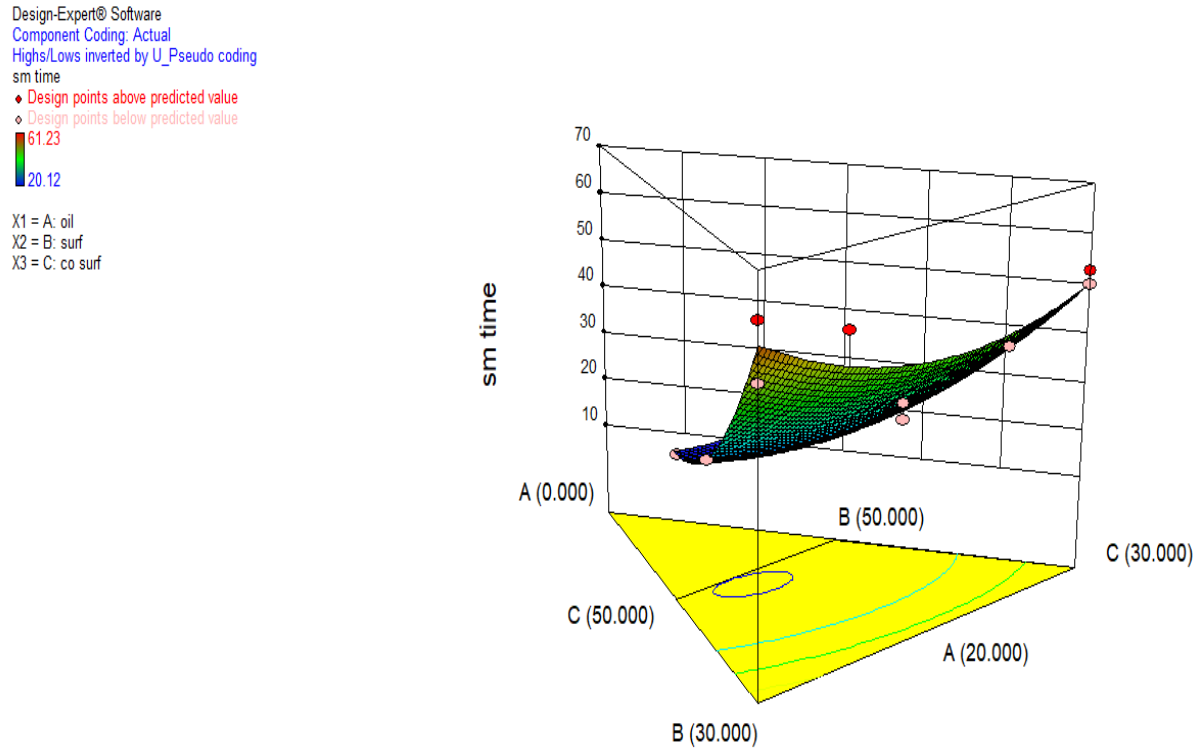


Fig 14. 3D Plot for Response 1 (Self emulsification time)

Response2: Globule Size Transform: None

Table 21. Sequential Model Sum of Squares [Type I]

Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Mean vs Total	503287.4	1	503287.4			
Linear vs Mean	1715.259	2	857.6296	45.09086	< 0.0001	Suggested
Quadratic vs Linear	118.7144	3	39.57145	3.078387	0.0772	
Sp Cubic vs Quadratic	4.198645	1	4.198645	0.303889	0.5949	
Cubic vs Sp Cubic	21.78394	3	7.261314	0.424789	0.7424	
Quadratic vs Cubic	37.33807	1	37.33807	2.862234	0.1515	Aliased
Residual	65.2254	5	13.04508			
Sp Quadratic vs Quadratic	34.36235	3	11.45412	0.851303	0.5087	
Quadratic vs Sp Quadratic	28.9583	2	14.47915	1.109932	0.3991	Aliased
Residual	65.2254	5	13.04508			
Total	505249.9	16	31578.12			

"Sequential Model Sum of Squares [Type I]": Select the highest order polynomial where the additional terms are significant and the model is not aliased.

Table 22. Model Summary Statistics

Source	Std. Dev.	R-Squared	Adjusted R-Squared	Predicted R-Squared	PRESS	
Linear	4.361196	0.874009	0.854625	0.805728	381.2627	Suggested
Quadratic	3.585332	0.934499	0.901749	0.849805	294.7603	
Special Cubic	3.717039	0.936639	0.894398	0.792352	407.5127	
Cubic	4.134478	0.947739	0.869347	-121.365	240142.8	
Special Quadratic	3.668081	0.952009	0.897162	0.558757	865.9483	
Quadratic	3.611797	0.966764	0.900293	+	Aliased	

+ Case(s) with leverage of 1.0000: PRESS statistic not defined I+"Model Summary Statistics"0+: Focus on the model maximizing the "Adjusted R-Square and the "Predicted R-Squared".

ANOVA for Response Surface Quadratic Model

Table 23. Analysis of variance table [Partial sum of squares - Type III]

Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	1715.259	2	857.6296	45.09086	< 0.0021	significant
Linear Mixture	1715.259	2	857.6296	45.09086	< 0.0001	
Residual	247.2604	13	19.02003			
Lack of Fit	182.035	8	22.75438	1.744288	0.2799	not significant
Pure Error	65.2254	5	13.04508			
Cor Total	1962.52	15				

The Model F-value of 45.09 implies the model is significant. There is only a 0.01% chance that a "Model F-Value" this large could occur due to noise.

Values of "Prob > F" less than 0.0500 indicate model terms are significant. In this case Linear Mixture Components are significant model terms. Values greater than 0.1000 indicate the model terms are not significant. If there are many insignificant model terms (not counting those required to support hierarchy), model reduction may improve your model.

The "Lack of Fit F-value" of 1.74 implies the Lack of Fit is not significant relative to the pure error. There is a 27.99% chance that a "Lack of Fit F-value" this large could occur due to noise. Non-significant lack of fit is good - we want the model to fit.

Std. Dev.	4.361196	R-Squared	0.874009
Mean	177.3569	Adj R-Squared	0.854625
C.V. %	2.458995	Pred R-Squared	0.805728
PRESS	381.2627	Adeq Precision	16.91413

The "Pred R-Squared" of 0.8057 is in reasonable agreement with the "Adj R-Squared" of 0.8546."Adeq Precision" measures the signal to noise ratio. A ratio greater than 4 is desirable. Ratio of 16.914 indicates an adequate signal. This model can be used to navigate the design space.

Component	Coefficient Estimate	df	Standard Error	95% CI Low	95% CI High	VIF
A-oil	128.7958	1	5.237398	117.4811	140.1105	1.460343
B-surf	190.3455	1	2.316757	185.3405	195.3506	1.317234
C-co surf	185.5835	1	2.264757	180.6908	190.4763	1.182077

Final Equation in Terms of U_Pseudo Components:

Globule size =+128.80 * A+190.35* B+185.58 * C

Final Equation in Terms of Real Components:

Globule size =+424.63947 * oil+116.89084* surf+140.70079 * co surf

Final Equation in Terms of Actual Components:

Globule size =+4.24639* oil+1.16891 * surf+1.40701 * co surf

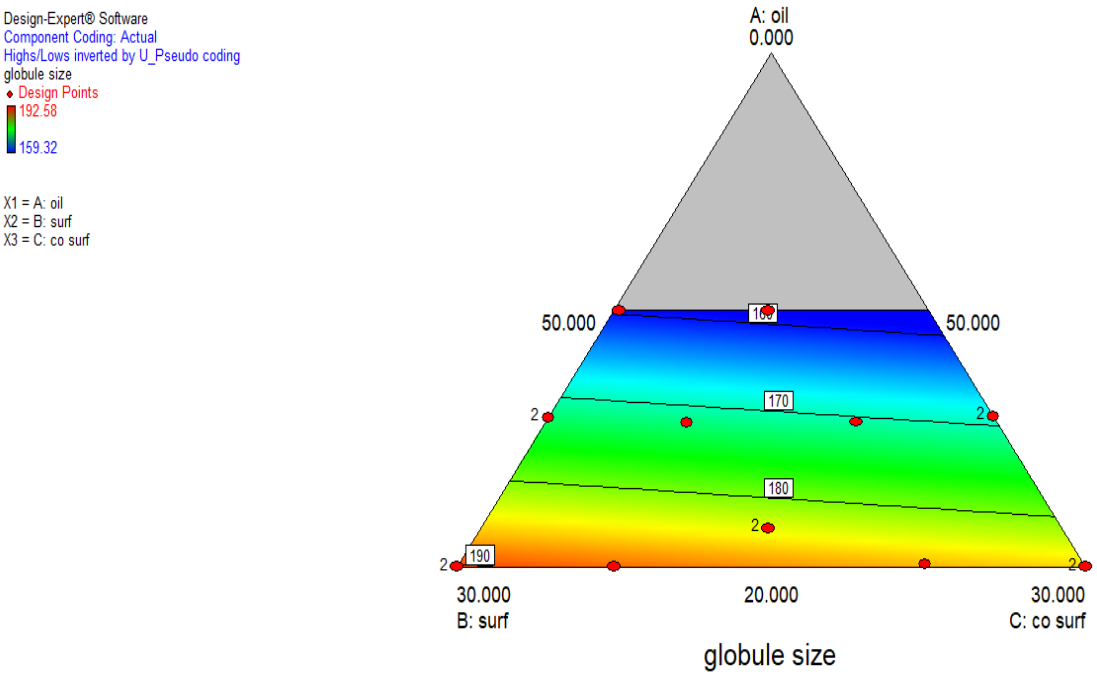


Fig 15. Contour Plot of Response 2 (Globule Size)

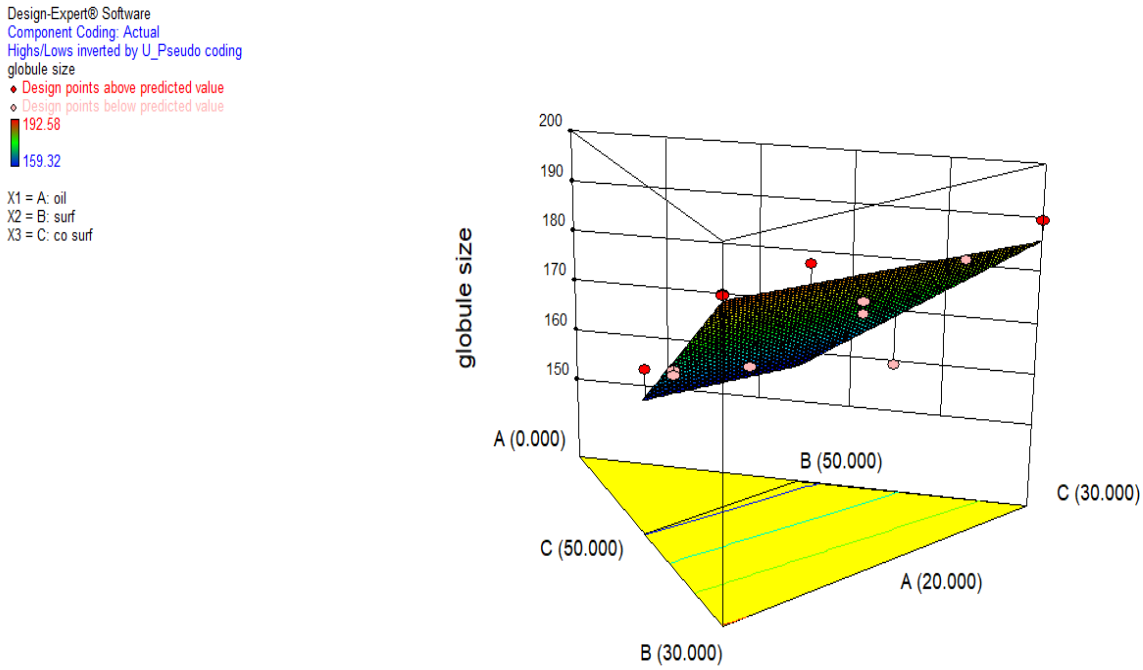


Fig 16. 3D Plot for Response 2 (Globule Size)

Response 3: % Transmittance Transform: None

Table 24. Sequential Model Sum of Squares [Type I]

Sum of Source	Mean Squares	df	p-value Square	Value	Prob > F	
Mean vs Total	153497.4	1	153497.4			
Linear vs Mean	11.5943	2	5.797151	27.83137	< 0.0001	
Quadratic vs Linear	2.20294	3	0.734313	14.54367	0.0006	Suggested
Sp Cubic vs Quadratic	0.027019	1	0.027019	0.508855	0.4937	
Cubic vs Sp Cubic	0.243983	3	0.081328	2.086213	0.2035	
Quadratic vs Cubic	2.35E-07	1	2.35E-07	5.01E-06	0.9983	Aliased
Residual	0.2339	5	0.04678			
Sp Quadratic vs Quadratic	0.172921	3	0.05764	1.215373	0.3726	
Quadratic vs Sp Quadratic	0.098082	2	0.049041	1.048329	0.4167	Aliased
Residual	0.2339	5	0.04678			
Total	153511.7	16	9594.484			

"Sequential Model Sum of Squares [Type I]": Select the highest order polynomial wherethe additional terms are significant and the model is not aliased.

Table 25. Model Summary Statistics

Source	Std. Dev.	R-Squared	Adjusted R-Squared	Predicted R-Squared	PRESS	
Linear	0.456394	0.810669	0.781541	0.695577	4.353895	
Quadratic	0.2247	0.964697	0.947046	0.911708	1.262759	Suggested
Special Cubic	0.23043	0.966587	0.944311	0.897862	1.460798	
Cubic	0.197442	0.983646	0.959114	0.936191	0.912609	
Special Quadratic	0.217775	0.976788	0.95026	0.794555	2.9383	
Quadratic	0.216287	0.983646	0.950937	+	Aliased	

+ Case(s) with leverage of 1.0000: PRESS statistic not defined *"Model Summary Statistics"*: Focus on the model maximizing the "Adjusted R Squared"and the "Predicted R-Squared".

ANOVA for Response Surface Quadratic Model

Table 26. Analysis of variance table [Partial sum of squares - Type III]

Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	13.79724	5	2.759448	54.65311	< 0.0061	significant
Linear Mixture	11.5943	2	5.797151	114.8173	< 0.0001	
AB	0.539875	1	0.539875	10.69267	0.0084	
AC	0.420402	1	0.420402	8.326396	0.0162	
BC	1.523117	1	1.523117	30.16657	0.0003	
Residual	0.504902	10	0.05049			
Lack of Fit	0.271002	5	0.0542	1.158625	0.4378	not significant
Pure Error	0.2339	5	0.04678			
Cor Total	14.30214	15				

The Model F-value of 54.65 implies the model is significant. There is only a 0.01% chance that a "Model F-Value" this large could occur due to noise.

Values of "Prob > F" less than 0.0500 indicate model terms are significant. In this case Linear Mixture Components, AB, AC, BC are significant model terms. Values greater than 0.1000 indicate the model terms are not significant. If there are many insignificant model terms (not counting those required to support hierarchy), model reduction may improve your model.

The "Lack of Fit F-value" of 1.16 implies the Lack of Fit is not significant relative to the pure error. There is a 43.78% chance that a "Lack of Fit F-value" this large could occur due to noise. Non-significant lack of fit is good -- we want the model to fit.

Std. Dev.	0.2247	**R-Squared**	0.964697
Mean	97.94688	**Adj R-Squared**	0.947046
C.V. %	0.22941	**Pred R-Squared**	0.911708
PRESS	1.262759	**Adeq Precision**	20.76396

The "Pred R-Squared" of 0.9117 is in reasonable agreement with the "Adj R-Squared" of 0.9470. "Adeq Precision" measures the signal to noise ratio. A ratio greater than 4 is desirable. Your ratio of 20.764 indicates an adequate signal. This model can be used to navigate the design space.

Component	Coefficient Estimate	df	Standard Error	95% CI Low	95% CI High	VIF
A-oil	-29.052	1	40.69659	-119.73	61.62561	14739.24
B-surf	70.86527	1	5.396423	58.84129	82.88925	1628.219
C-co surf	77.29584	1	4.931347	66.30811	88.28356	1404.947
AB	202.2783	1	61.85945	64.4468	340.1097	5893.256
AC	159.3364	1	55.21873	36.30138	282.3714	4764.794
BC	94.90586	1	17.27946	56.40483	133.4069	2786.754

Final Equation in Terms of Actual Components:

% transmittance =-0.29052 * oil+0.7086 * surf +0.77296 * co surf+0.020228 * oil * surf +0.015934 * oil * co surf +9.49059E-003 * surf * co surf

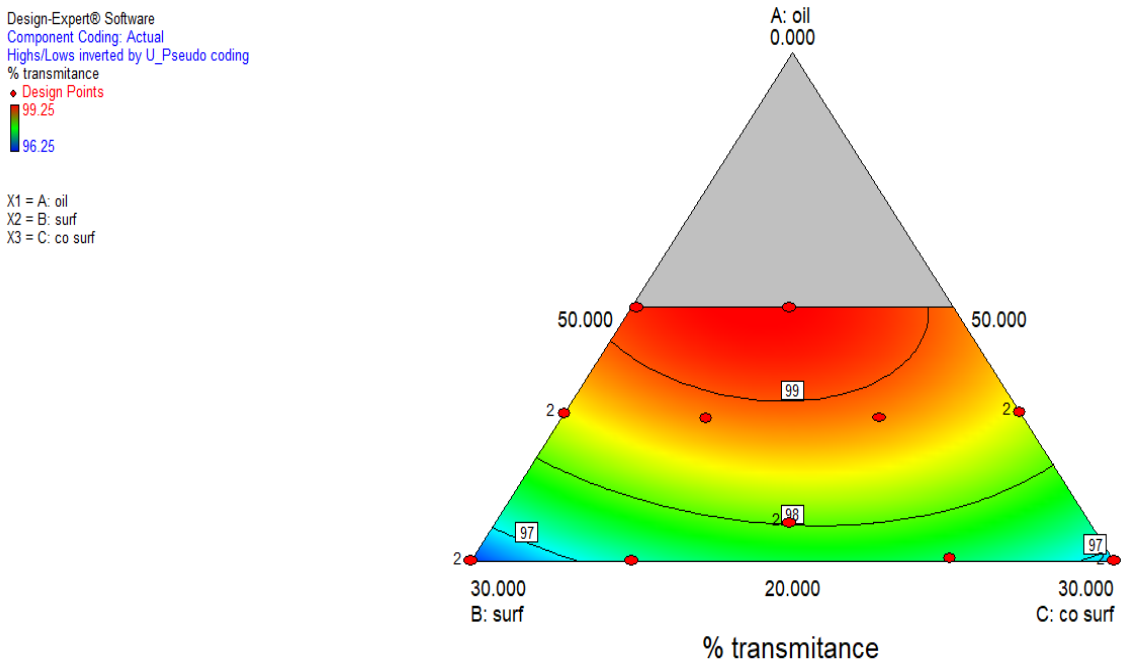


Fig 17. Contour plot for Response 3 (% Transmittance)

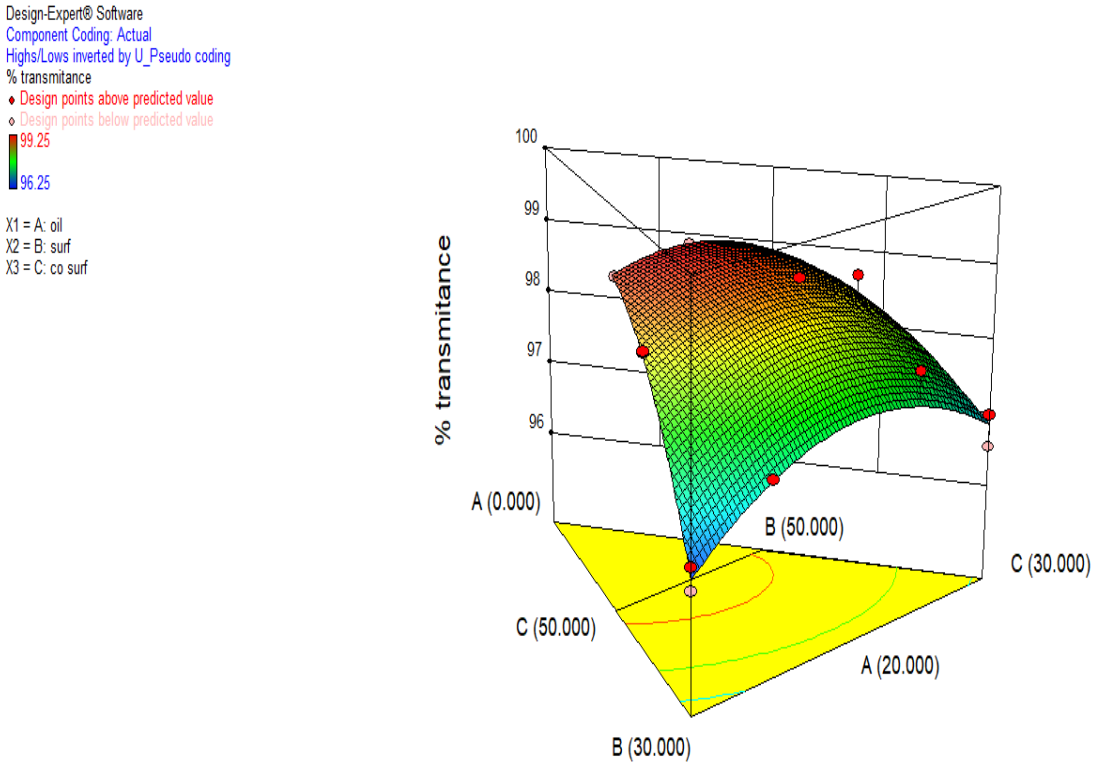


Fig 18. 3D plot for Response 3 (% Transmittance)

Fig 19. Linearity plot for Response 1(Self Emulsification time)

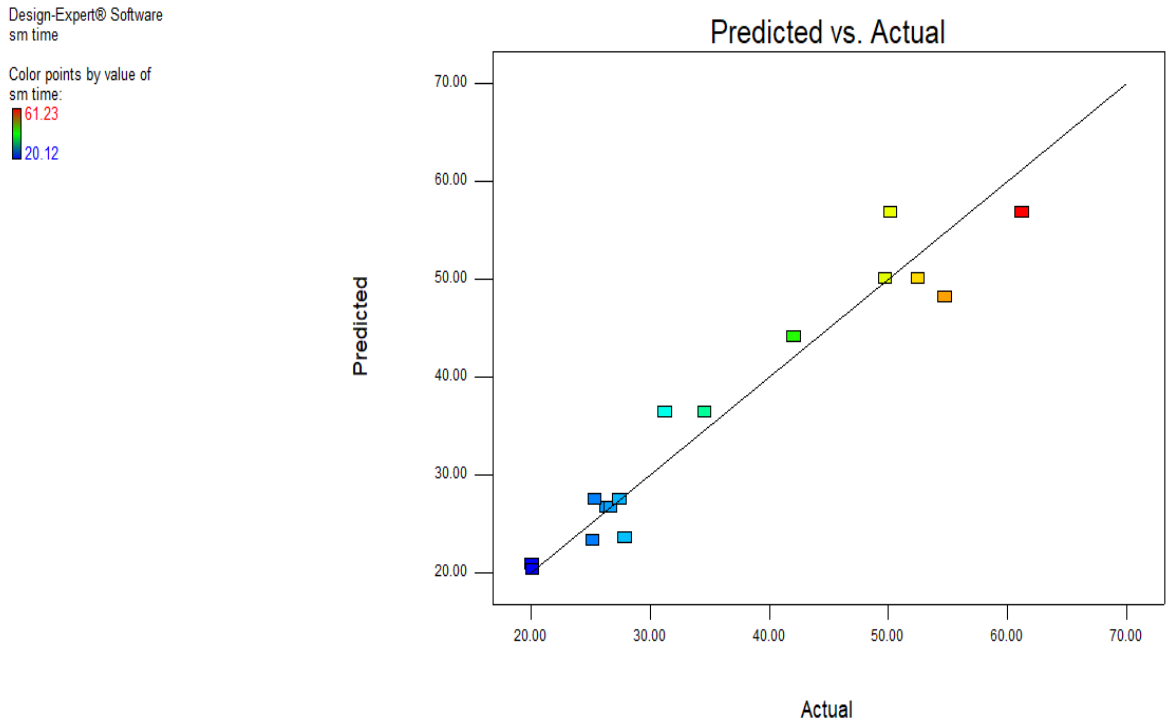
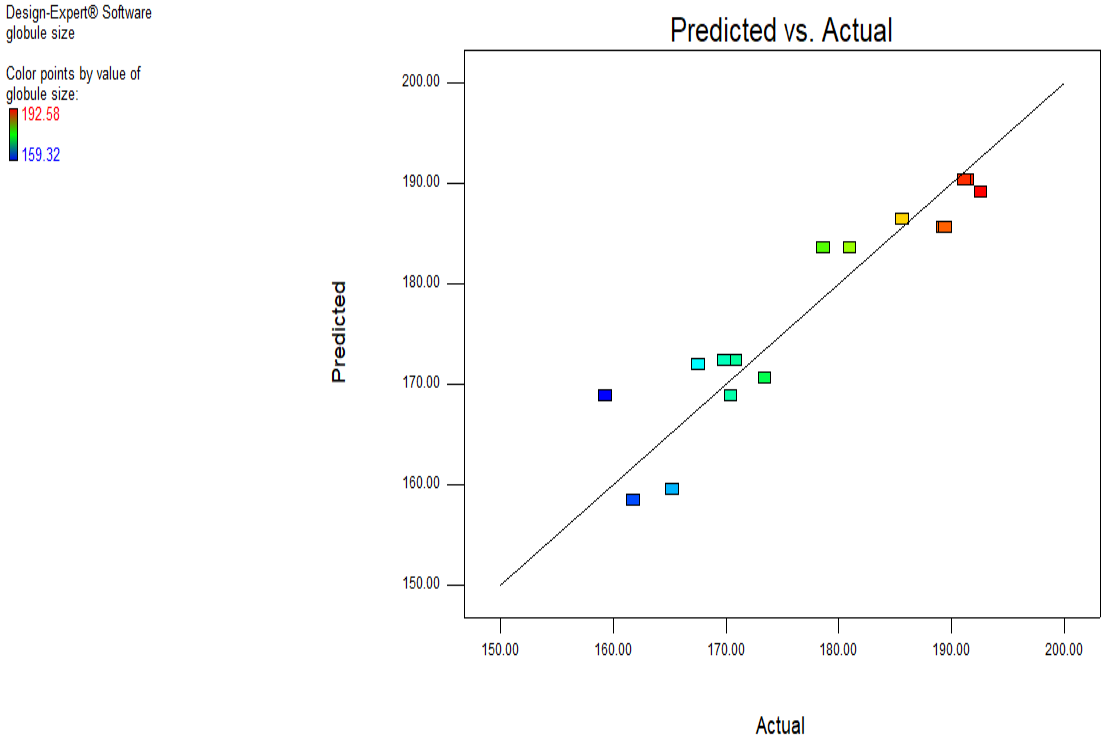


Fig 20. Linearity plot for Response 2 (Globule size)



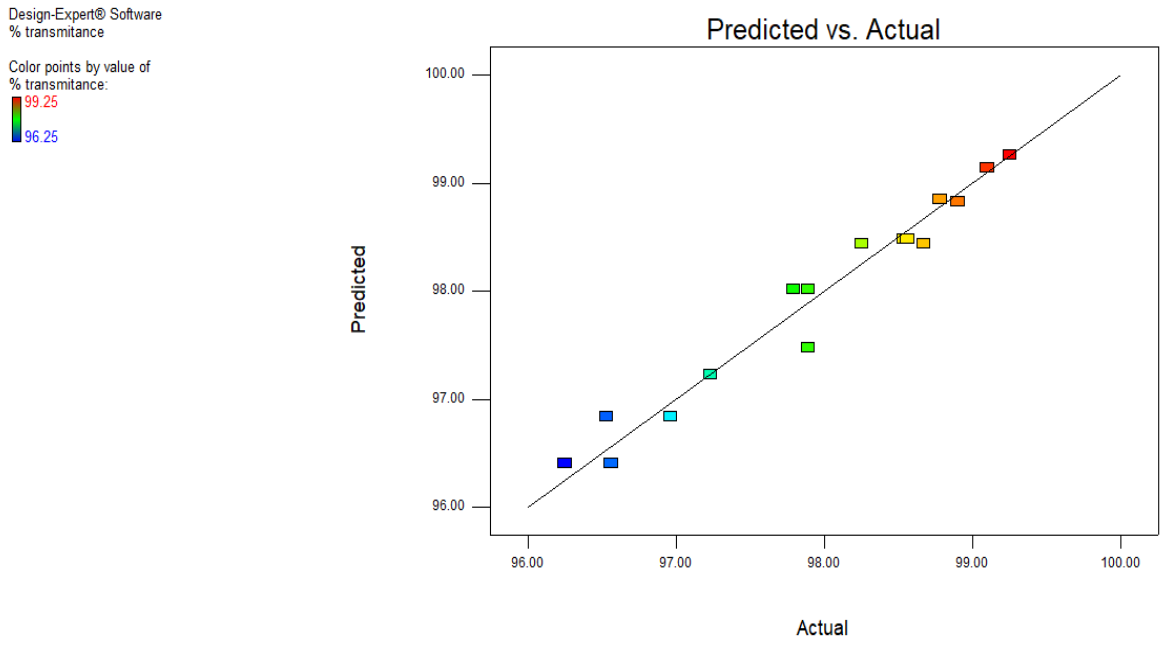


Fig 21. Linearity plot for Response 3(% Transmittance)

Table 27. Formulation Table for Optimized Formulation

Factor	Name	Level	Low Level	High Level	Coding
A	Oil	10.000051	10	20	Actual
B	Surfactant	48.00467	20	50	Actual
C	Co-Surfactant	42.474508	20	50	Actual

Table 28. Predicted values for Optimized formulation

Response	Prediction	Std Dev	SE Mean	95% CI low	95% CI high	SE Pred	95% PI low	95% PI high	95% TI low	95% TI high
s-m time	26.87149	4.183781	2.009246	22.39462	31.34837	4.641238	16.53017	37.21282	6.061749	47.68124
globule size	176.1308	4.361196	1.098878	173.7568	178.5048	4.497506	166.4145	185.8471	157.4982	194.7634
% transmittance	98.62124	0.2247	0.107912	98.3808	98.86168	0.249269	98.06583	99.17665	97.5036	99.73888

D- Optimal design predicted formulation F4 may be an optimized formulation.
On other hand F10 also were in the Range of good formulation.

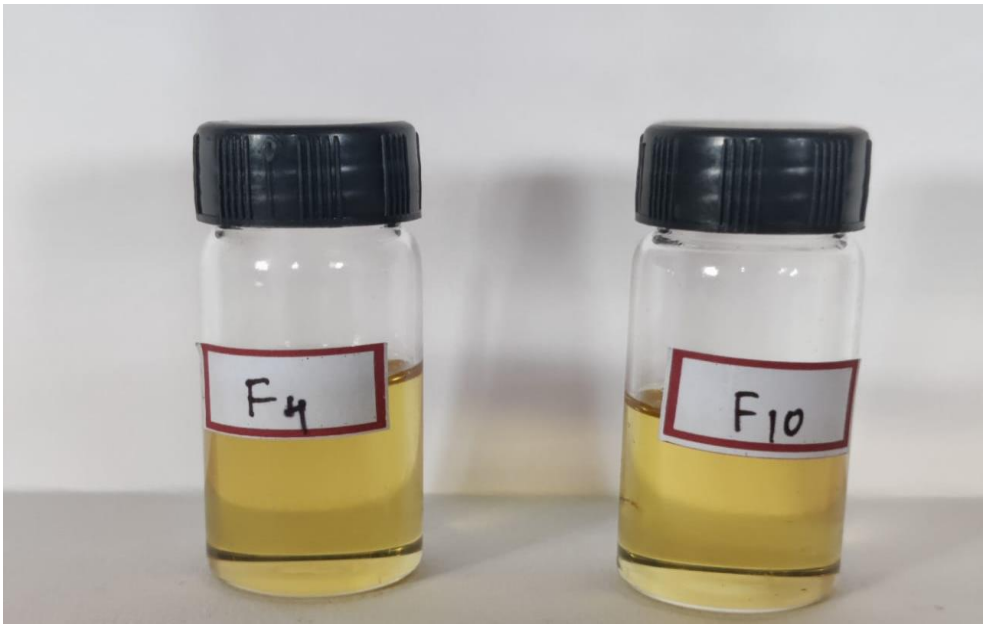
Table 29. Comparison of Predicted and Actual values of Optimized formulation

Parameters	Predicted Value	Actual Value	SE Mean
Self Emulsification Time (Sec)	26.87149	20.54	1.95075
Globule Size (nm)	176.1308	158.2	3.06090
% Transmittance	98.62124	99.25	0.35245

FORMULATION OF SELF MICRO EMULSIFYING DRUG DELIVERY SYSTEM(SMEDDS)

Series of Liquid self micro emulsifying drug delivery system (L-SMEDDS) were prepared by vortex method using various concentrations of Oil, Surfactant and Co-surfactant with Ivermectin.

Fig.22 L-SMEDDS Formulation



CHARACTERIZATION OF L-SMEDDS FORMULATIONS

Determination of Self-Emulsification Time

The efficiency of self-emulsifying system was assessed from the rate of emulsification upon hydration with mild agitation. Surfactant system in SMEDDS formulation reduces the interfacial tension between oil and aqueous phase resulting in easy dispersion and formation of oil-in-water (O/W) emulsion. The self-emulsification time of all L-SMEDDS formulations are presented in Table 28.

Table 30. Self-Emulsification Time of L-SMEDDS Formulations

S.No.	Formulation code	Self-emulsification time (sec)
1	F1	42.09
2	F2	25.36
3	F3	27.89
4	F4	20.15
5	F5	26.35
6	F6	52.48
7	F7	34.58
8	F8	49.75
9	F9	50.19
10	F10	21.12
11	F11	31.25
12	F12	25.19
13	F13	61.23
14	F14	26.69
15	F15	27.45
16	F16	54.76

With the increase in oil proportion there was a decrease in the rate of emulsification and increase in emulsification time. The higher interfacial tension between larger volume of oil and aqueous phase and decrease in concentration of surfactant system may be responsible for

the increased emulsification time. The F4 and F10 formulation has taken short time (20.15 and 21.12 seconds respectively) to emulsify and F13 formulation has taken long time (61.23 seconds) to emulsify.

Determination of % Transmittance

The clarity of Micro emulsion can be observed by transparency, which can be measured in the form of % transmittance (% T). The prepared L-SMEDDS formulations are O/W type of nanoemulsion. The % transmittance above 90% indicates that the formulations are transparent. The results of % transmittance study are shown in Table 29.

Table 31. % Transmittance Study of L-SMEDDS Formulations

S.No.	Formulation code	% Transmittance
1	F1	97.89
2	F2	98.25
3	F3	98.9
4	F4	99.25
5	F5	98.54
6	F6	96.96
7	F7	97.79
8	F8	96.53
9	F9	96.25
10	F10	99.1
11	F11	97.89
12	F12	98.78
13	F13	96.56
14	F14	98.56
15	F15	98.67
16	F16	97.23

All the prepared formulations have good % transmittance and they are transparent. The lower values of % transmittance could be due to larger globule size of prepared nanoemulsions. The formulation F4 shows 99.25 % of transmittance.

Determination of Particle size

Smaller globule size of the emulsion droplets may lead to more rapid absorption and improved bioavailability. The Polydispersity index is also an important parameter. It gives the size distribution of globules and uniform distribution of L-SMEDDS in aqueous medium. The results of Particle size study are presented in the Table 30.

Table 32. Particle size Study of L-SMEDDS Formulations

S.No.	Formulation code	Particle size
1	F1	185.65
2	F2	170.45
3	F3	173.45
4	F4	161.78
5	F5	170.87
6	F6	189.45
7	F7	180.98
8	F8	189.26
9	F9	191.45
10	F10	165.25
11	F11	178.65
12	F12	167.58
13	F13	191.12
14	F14	169.87
15	F15	159.32
16	F16	192.58

The prepared formulations have the globule size range between 161-192nm.

Determination of Refractive Index (RI)

The clarity of micro emulsion is estimated by measuring the refractive index of the L-SNEDDS formulations. In the present study the refractive index of distilled water (standard) was used for comparison with the prepared formulations. The results of RI study are presented in the Table 33.

Table 33. Refractive Index Study of L-SNEDDS Formulations.

S.No.	Formulation code	Refractive Index (RI)
1	F1	1.33390

The RI of water is 1.333250 at 25 ˚ C (Atago Rx-7000i). If the formulations show the same value of RI the formulations are clear. The RI values of all formulations were closed to Distilled water. So, the formulations were clear as water (indication of formation of nanoemulsion).

Determination of pH of L-SMEDDS

Stability of SMEDDS formulations could be greatly affected by pH. The change in the pH might affect the Zeta potential of the formulations which in turn affect the stability of preparations. The results for the pH study are presented in Table 31.

Table 34. pH of L-SMEDDS Formulations

S.No.	Formulation Code	pH*
1	F1	6.68 ± 0.03

All the prepared formulations showed similar pH values in the range of 6.6 to 6.9. It can be assumed that drug is not diffusing in the external phase and remains in the oil phase.

Drug Content Study of L-SMEDDS

The UV-visible spectrophotometric method was used to determine the drug content of Ivermectin L-SMEDDS formulations. Drug content of all the L-SMEDDS formulations are presented in Table 32.

Table 35. Drug Content of L-SMEDDS Formulations

S.No.	Formulation Code	Drug Content(%w/w)
1	F1	99.54

Drug content of the prepared formulations was within the specified limit (90.0% - 110.0% as per the IP).

Phase Separation and Precipitation Study

Phase separation study was assessed by exposing SMEDDS formulations to 100 fold dilution with 0.1 N HCl. The formulations were stored at 25^0C for 48 hours and observed visually for phase separation and precipitation of drug. The results are presentedin Table 33.

Table 36. Phase Separation and Precipitation of Drug from L-SMEDDS Formulations

S.No.	Formulation Code	Phase Separation	Drug Precipitation
1	F1	No	No

Phase separation studies after 100 times dilution with 0.1 N HCl reveals that formulations F1 are stable for a period of 48 hrs. There is no phase separation and precipitation of drug.

Thermodynamic stability studies

The physical stability of the formulation is very important for its performance as it can be adversely affected by precipitation of the drug in an excipient matrix. Poor physical stability of the formulation can lead to phase separation of excipients that affects bioavailability, as well as therapeutic efficacy. Furthermore, the incompatibility between formulation and gelatin shell caused brittleness, softness and delayed the disintegration or incomplete release of the drug.

Table 37. Thermodynamic stability study of L-SMEDDS Formulations

Formulation Code	Observations based on the thermodynamic stability tests			Inference
	Heating cooling cycle	Centrifugation study	Freeze thaw	
F1	✓	✓	✓	Passed

It was observed that, there were no appreciable change in the formulations F1 during stability studies, and hence it was concluded that the formulation are thermodynamically stable.

Dispersibility Test for L-SMEDDS Formulation

The stability of L- SMEDDS formulation was estimated by dispersibility test. The L-SMEDDS formulations were distributed to various media like Water (Neutral media), 0.1 N HCl (Strong acidic media), and Phosphate buffer pH 6.8 (Weak acidic media). The results are shown in Table 35.

Table 38. Dispersibility study of L-SMEDDS Formulations.

Formulation Code	Observations based on the Dispersibility tests			Inference
	Distilled water	0.1 N HCl	Phosphate buffer pH 6.8	
F1	Grade A	Grade A	Grade A	Passed

The formulation F1 passed the Dispersibility test on various media. Hence these formulations show same Grade of emulsion and same degree of stability in all media (0.1 N HCl, water, Phosphate buffer pH6.8)

Determination of viscosity

Viscosity Determination Before dilution

The viscosity of the prepared nanoemulsion formulations were determined as such without dilution by Brookfield DV III ultra V6.0 RV cone and plate rheometer (Brookfield Engineering Laboratories, Inc, Middleboro, MA) using spindle # CPE40 at 25 ±0.5°C. The software used for the viscosity calculations was Rheocalc V2.6.

Viscosity Determination After dilution

The viscosity of diluted SNEDDS was determined by Malvern Zeta sizer. The viscosity of Diluted SNEDDS shows the type of emulsion formed. Low viscosity indicates the formation of **w/o** emulsion and high viscosity indicates **o/w** emulsion.

Table 39. Viscosity Determination for L- SNEDDS Formulations

Formulation Code	Viscosity Before dilution in cps*	Viscosity After dilution in cps
F1	84.0 ± 0.25	0.8872

Zeta Potential Analysis

Smaller globule size of the emulsion droplets may lead to more rapid absorption and improved bioavailability. The Polydispersity index is also an important parameter. It gives the size distribution of globules and uniform distribution of L-SMEDDS in aqueous medium. If the value is less than 0.2 the emulsions has good dispersion. The degree of attraction or repulsion between globules could be measured by zeta potential. The globules present in the polar medium like water would possess a surface charge.

The blank SMEDDS formulation exhibited almost no charge whereas a negative charge was obtained with drug loaded SMEDDS. This could be because of the emulsifier used in the formulations. The higher the zeta potential, greater will be the energy barrier to coalescence between oil globules and so higher will be the stability. The study results are presented in the Table 40.

Table 40. Zeta Potential of L-SMEDDS Formulations

S.No.	Formulation code	Zeta potential (mV)	Polydispersity index (PDI)
1	**F1**	**-0.3**	**0.087**

Fig 23. Size Distribution of F1 formulation.

HORIBA Scientific

SZ-100

HORIBA SZ-100 for Windows [Z Type] Ver2.20

Measurement Results

Date	: 15 February 2022 11:32:08
Measurement Type	: Particle Size
Sample Name	: Smedds
Scattering Angle	: 90
Temperature of the Holder	: 24.9 deg. C
Dispersion Medium Viscosity	: 0.897 mPa.s
Transmission Intensity before Meas.	: 11525
Distribution Form	: |Standard|
Distribution Form(Dispersity)	: Monodisperse
Representation of Result	: Scattering Light Intensity
Count Rate	: 2029 kCPS

Calculation Results

Peak No.	S.P.Area Ratio	Mean	S. D.	Mode
1	1.00	158.3 nm	40.9 nm	160.3 nm
2	---	--- nm	--- nm	--- nm
3	---	--- nm	--- nm	--- nm
Total	1.00	158.3 nm	40.9 nm	160.3 nm

Cumulant Operations

Z-Average	: 158.3 nm
PI	: 0.372

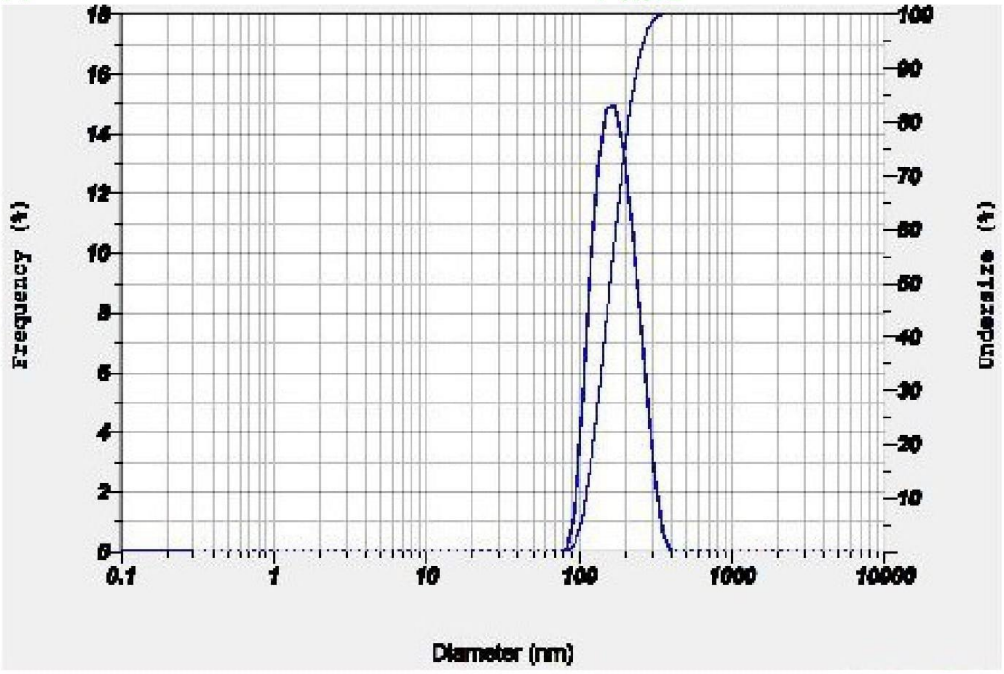


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Fig 24. Zeta Potential of F1 Formulation.

HORIBA Scientific

HORIBA SZ-100 for Windows [Z Type] Ver2.20

SZ-100

Measurement Results

Measurement Results

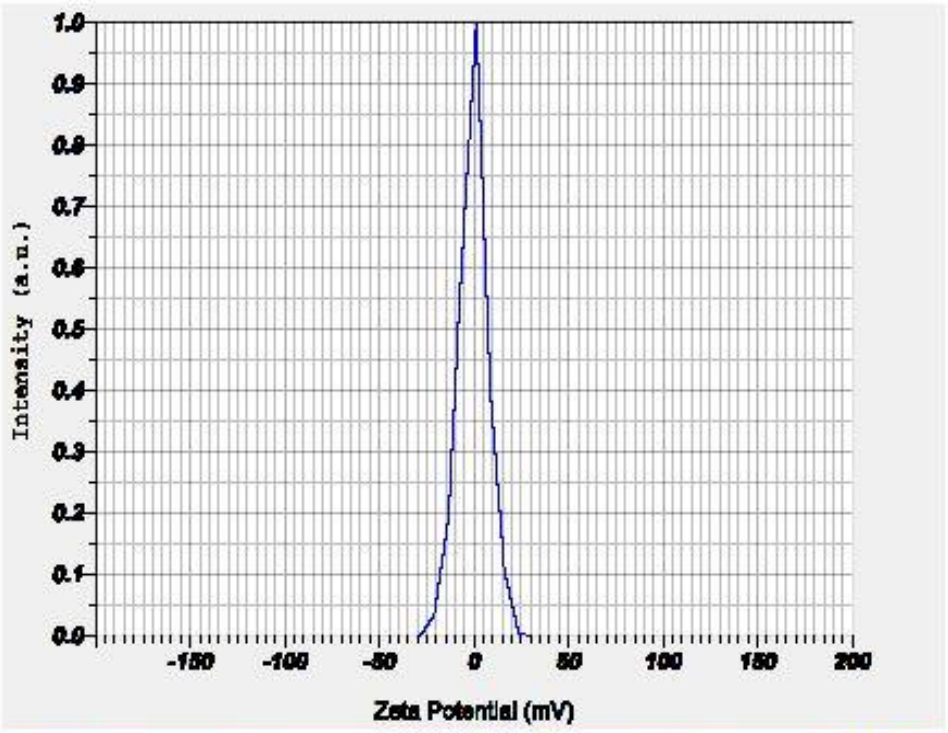
Date	: 15 February 2022 12:10:47
Measurement Type	: Zeta Potential
Sample Name	: SMEDDS 15022022
Temperature of the Holder	: 25.0 deg. C
Dispersion Medium Viscosity	: 0.894 mPa.s
Conductivity	: 0.245 mS/cm
Electrode Voltage	: 3.4 V

Calculation Results

Peak No.	Zeta Potential	Electrophoretic Mobility
1	-0.3 mV	-0.000002 cm2/Vs
2	— mV	— cm2/Vs
3	— mV	— cm2/Vs

Zeta Potential (Mean) : -0.3 mV

Electrophoretic Mobility Mean : -0.000002 cm²/Vs



HORIBA

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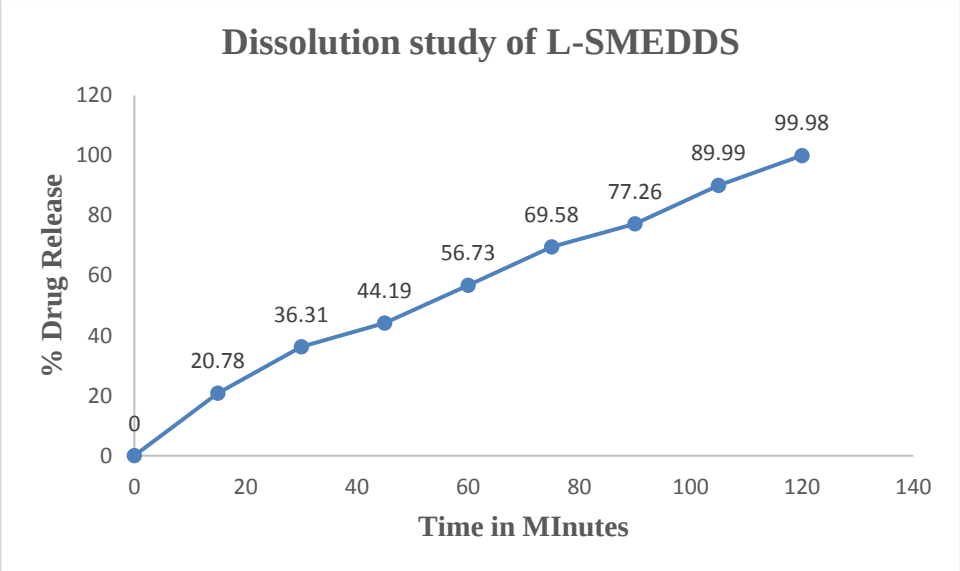
In Vitro Drug Release Study of L-SMEDDS

The dissolution of Ivermectin L-SMEDDS formulation in 0.1 N HCl (pH-1.2) was compared with pure drug powder and marketed formulation. The results are presented in Table 37. Fig 23.

Table 41. *In Vitro* Drug Release Study of L-SMEDDS

S.No.	Time (min)	% Cumulative Drug Release
1	15	20.78
2	30	36.61
3	45	44.19
4	60	56.73
5	75	69.58
6	90	77.26
7	105	89.99
8	120	99.98

Fig.25.*In Vitro* Drug Release Study of L-SMEDDS



In Vitro Drug Release Study of Marketed formulation

The dissolution of Ivermectin SMEDDS formulation in 0.1 N HCl (pH-1.2) was compared with pure drug powder and marketed formulation. The results are presented in Table 38. Fig 24**.**

Table 42. *In Vitro* Drug Release Study of Marketed Formulation

S.No.	Time (min)	% Cumulative drug release
1	15	11.27
2	30	23.34
3	45	27.69
4	60	39.91
5	75	45.20
6	90	59.71
7	105	66.82
8	120	75.26
9	135	88.77
10	150	93.56
11	165	99.25
12	180	102.04

Fig.26.*In Vitro* Drug Release Study of Marketed Formulation

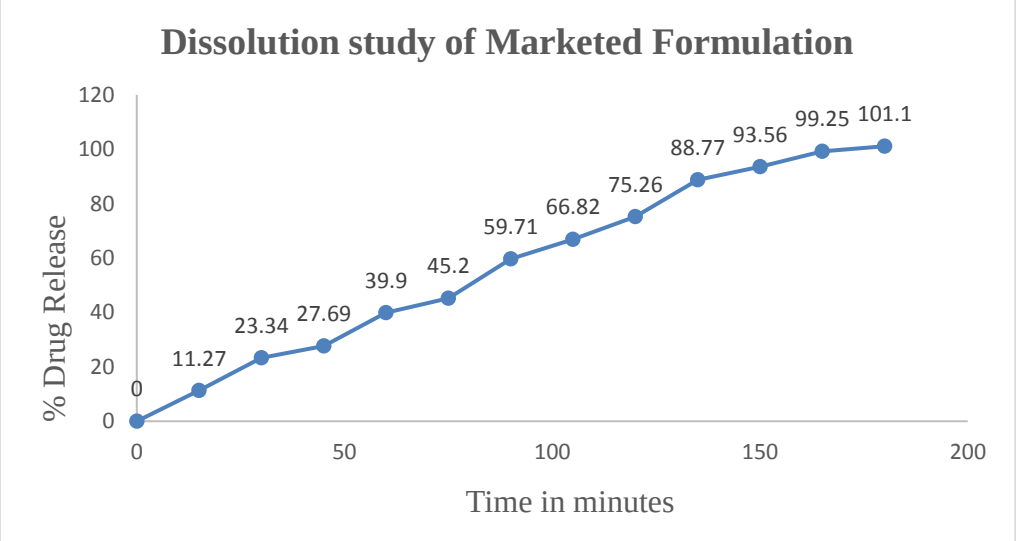
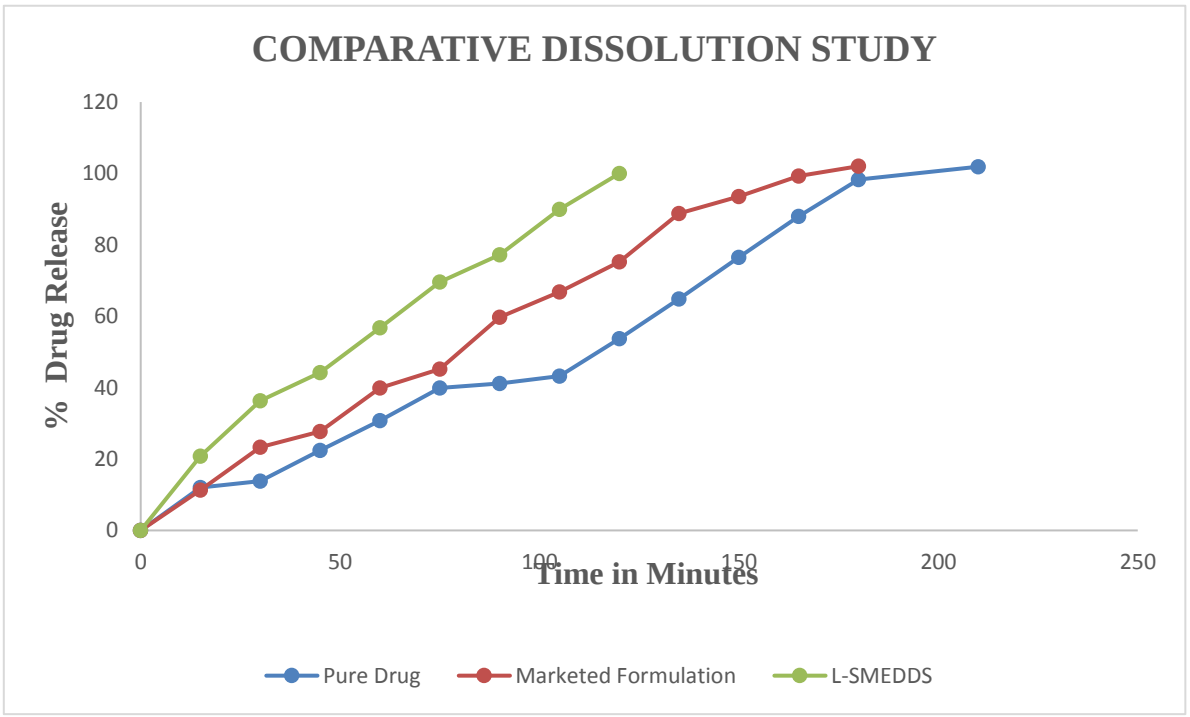


Table 43. Comparison of *In Vitro* Drug Release of Pure Drug, L-SMEDDS and Marketed Formulation

Time (min)	% Drug release		
	Pure drug	L-SMEDDS	Marketed Drug
0	0	0	0
15	12.05	20.78	11.27
30	13.84	36.61	23.34
45	22.39	44.19	27.69
60	30.76	56.73	39.91
75	39.91	69.58	45.2
90	41.15	77.26	59.71
105	43.2	89.99	66.82
120	53.71	99.98	75.26
135	64.82		88.77
150	76.5		93.56
165	87.97		99.25
180	98.26		102.04
210	101.89		

Fig. 27.Comparison of *In Vitro* Drug Release of Pure Drug, L-SMEDDS and Marketed Formulation



IN VITRO RELEASE KINETICS OF L-SMEDDS FORMULATION.

The values obtained from *in vitro* dissolution of L-SMEDDS formulation of Ivermectin were fitted in various kinetic models. The results are presented in Table 44. and Fig 28, 29, 30.31 and 32.

Table 44. *In vitro* release kinetics of L-SMEDDS formulation.

Time in mins	% Drug release	% Drug remaining	Log % cumulative Drug remaining	Log time	Log %Drug remaining	Cube Root % Drug Remaining	Square Root of Time
0	0	100	2	∞	∞	4.6415	0
15	20.78	79.22	1.8988	1.176	1.3176	4.2948	0.5
30	36.31	63.99	1.804	1.477	1.56	3.9935	0.7071
45	44.19	55.81	1.7467	1.653	1.6453	3.8215	0.866
60	56.73	43.27	1.6361	1.778	1.7538	3.5107	1
75	69.58	30.42	1.4831	1.875	1.8424	3.1216	1.118
90	77.26	22.74	1.3567	1.954	1.8876	2.8331	1.2247
105	89.99	10.01	1.0004	1.243	1.9541	2.1551	1.3228
120	99.98	0.02	-1.6989	1.301	1.9999	0.271	1.4142

Fig 28. Zero Order kinetics

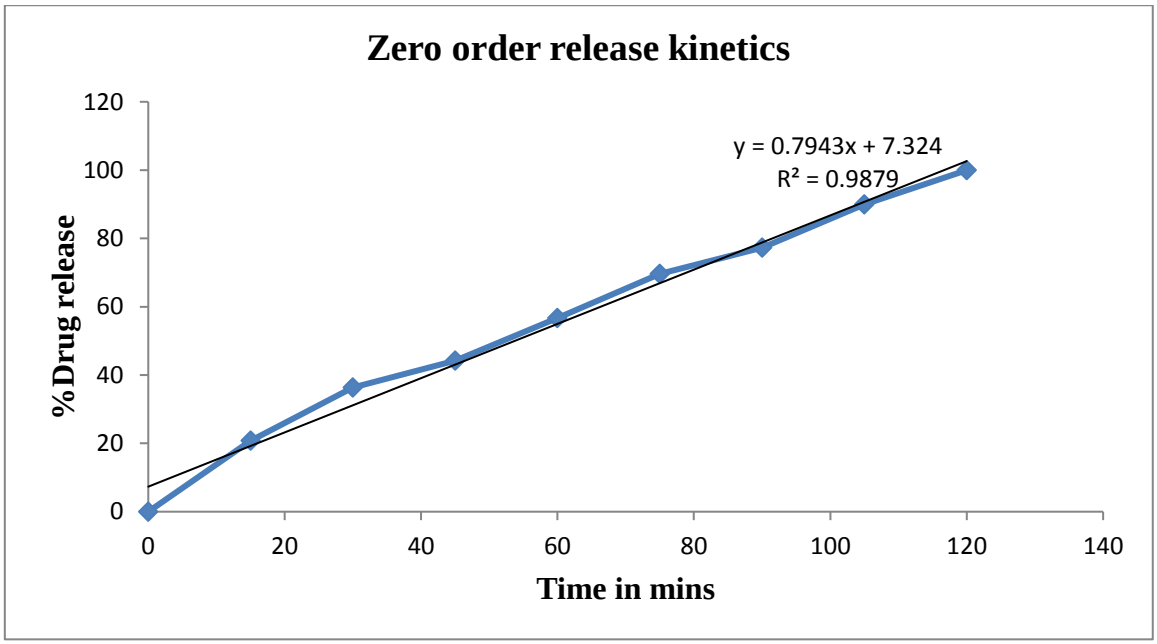


Fig 29. First order kinetics

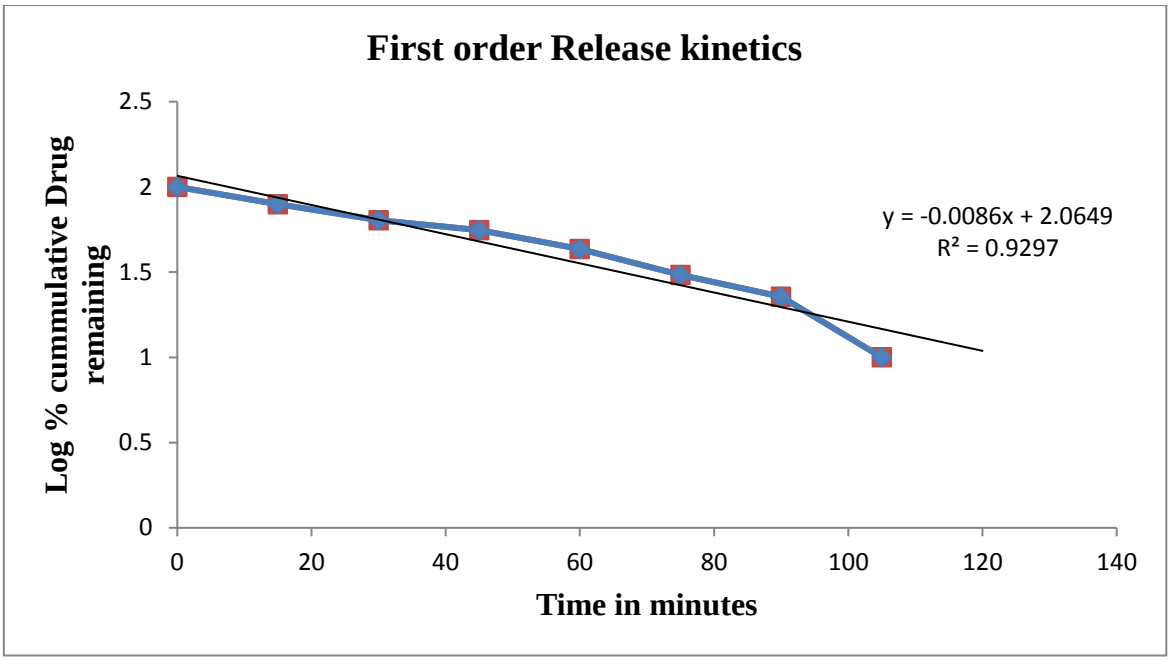


Fig.30.Higuchi diffusion kinetics

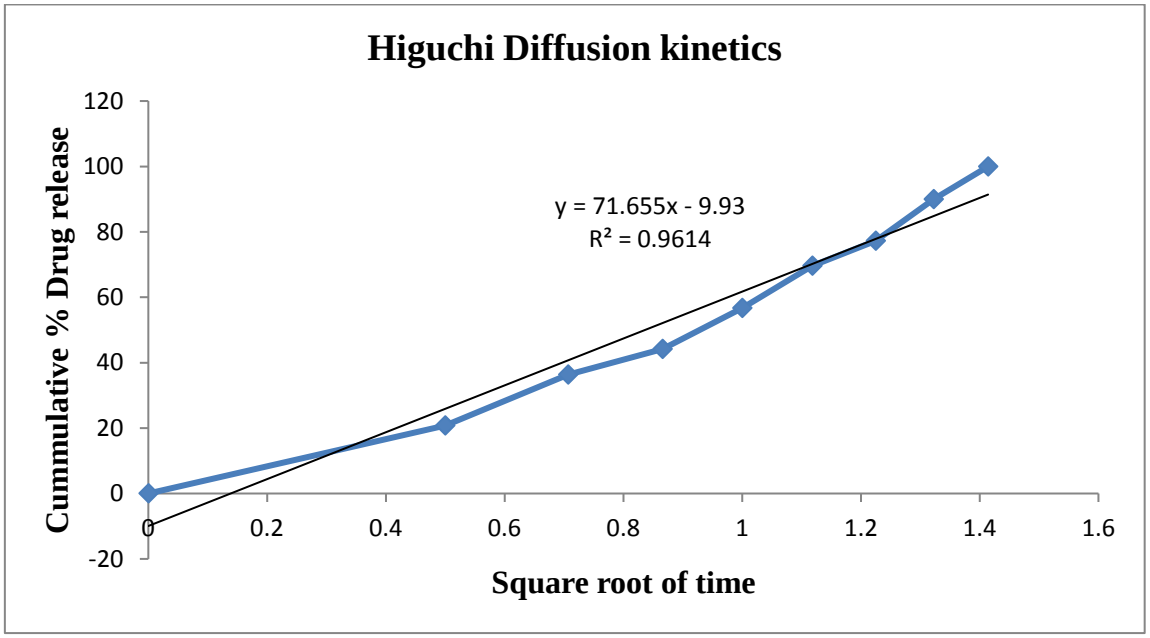


Fig.31 Hixson and Crowell release kinetics

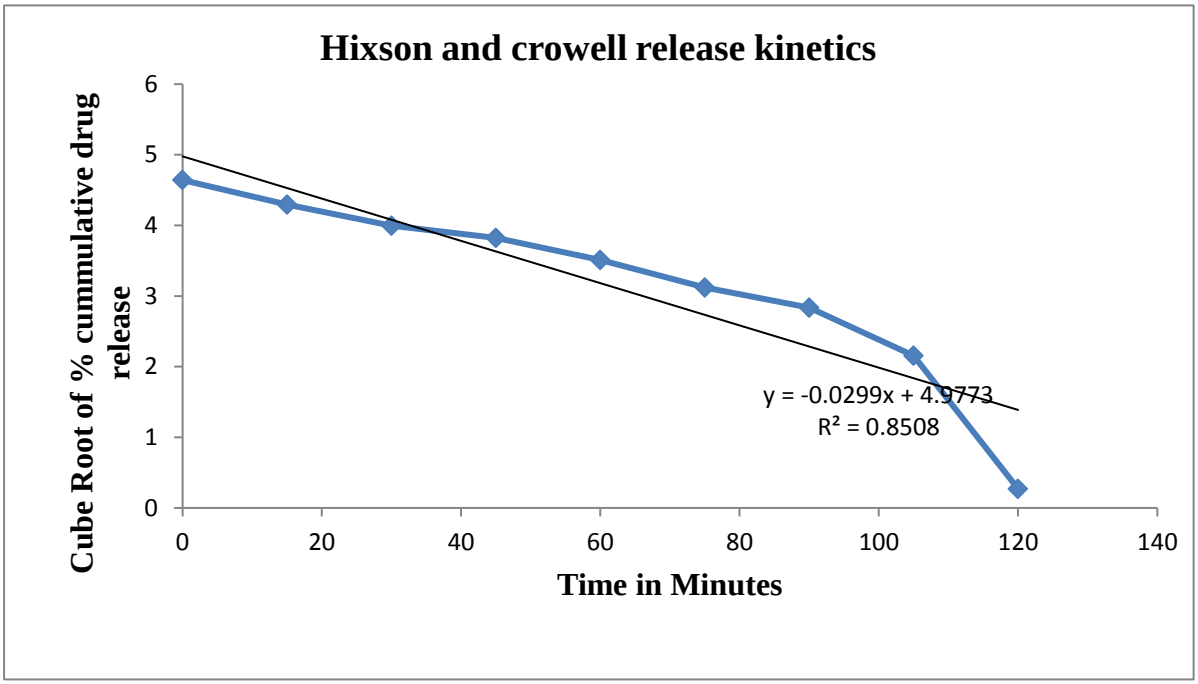
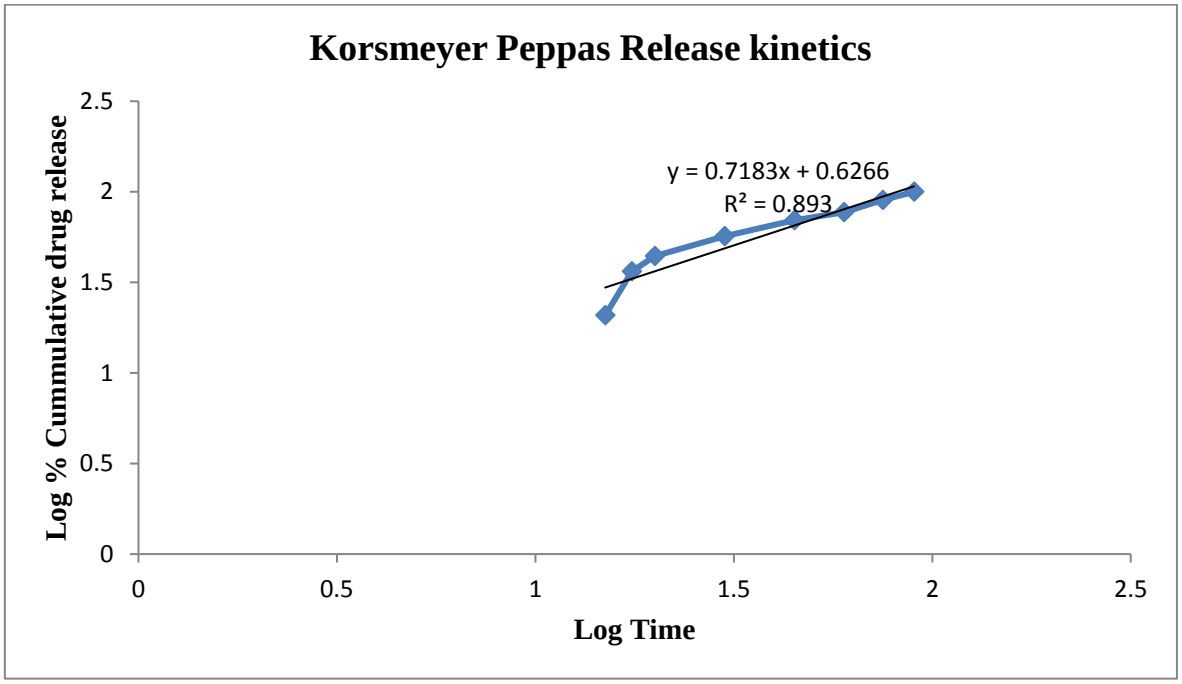


Fig. 32.Korsmeyer peppas release kinetics



The order of drug release was found to be Zero order, in which R^2 value was close to 1 than R^2 value of the first order equation. The slope of Higuchi equation was more than one, which indicates it follows Higuchi kinetics of release. The 'n' value, an exponent of Korsemeyer-Peppas equation was 0.718 indicating that the mass transfer follows Non-Fickian diffusion. As there are significant differences in the R^2 values of Zero order equation and Hixson-Crowell equation indicates the release by erosion mechanism.

ACCELERATED STABILITY STUDY

In order to evaluate the stability of the optimized SMEDDS, the formulation was added into sealed glass vials and the vial were subjected to stability studies at 40 ˚ C ± 2 ˚ C/ 75% ± 5% RH for a period of 3 months. Samples were charged in stability chambers (Thermo lab) with humidity and temperature control. The samples were evaluated for clarity, phase separation, precipitation, drug content and pH.

Table 45. Accelerated Stability study of L- SNEDDS

Testing period	Changing of Physical appearance		pH	Drug content (%w/w)	Phase separation	Precipitation
	Color	Odor				
Day 0	NC	NC	6.84	101.53	NO	NO
Day 15	NC	NC	6.86	101.53	NO	NO
Day 30	NC	NC	6.84	100.50	NO	NO
Day 60	NC	NC	6.88	99.90	NO	NO
Day 90	NC	NC	6.90	98.50	NO	NO

SUMMARY AND CONCLUSION

10. SUMMARY AND CONCLUSION

- The number of potential drug candidates that have poor aqueous solubility is progressively increasing. Accordingly, the problem has become dominant in the pharmaceutical industry.
- Formulation plays a major role in determining the rate and extent of absorption of such drugs from GIT. There are a number of drug strategies that could be used to improve the bioavailability of these drugs, either by increasing the dissolution rate or by presenting the drug in solution in the intestinal lumen. But most of the techniques used were with limitation of reproducibility, and stability.
- Among different approaches that have developed to enhance the aqueous solubility, the use of the self-Micro emulsifying drug delivery systems (SMEDDS) has drawn considerable attention and ultimately therapeutical and commercial success in the oral delivery of Poorly water soluble drug delivery system (PWSDs).
- They provide the PWSDs in the form of solubilized microdispersions. Consequently, the rate-limiting step of the PWSDs dissolution is bypassed. Nonetheless, SMEDDS are typically filled in soft gelatin capsules, which might cause the following problems: interaction with the capsule shell, instability, higher production cost and possible drug precipitation. Therefore, alternative formulation strategies, e.g. the inclusion of SMEDDS into a solid or semisolid dosage form, are desirable; nevertheless, very challenging.
- These formulations can also enhance drug absorption by a number of ancillary mechanisms, including inhibition of P-glycoprotein- mediated drug efflux, inhibition of pre-absorptive metabolism by gut membrane-bound cytochrome enzymes and/or promotion of lymphatic transport, which delivers drug directly to the systemic circulation while avoiding hepatic first-pass metabolism, and by increasing GI membrane permeability.
- Self-Micro emulsifying drug delivery system usually includes oil, non-ionic surfactant and co-surfactant, and the performance of these formulations depends upon the right combination of these.

- Most of the oils have the limitation of drug solubility and thus large amount of surfactant and co-surfactant are incorporated in the SMEDDS formulation to solubilize the drug.
- Toxicity of these surfactants is an independent issue, and is important with regard to the choice of surfactants so it is pertinent to compare the toxicity of non-ionic surfactants.
- Co- surfactant e.g. ethanol, propylene glycol, these alcohols and other volatile co-solvents have the disadvantage of evaporation into the shells of the soft gelatin capsules in conventional SMEDDS leading to drug precipitation.
- The absolute bioavailability of Ivermectin is approximately 56 %. Poor aqueous solubility, substrate to efflux mechanism of the GIT, high intestinal clearance and first-pass metabolism, are thought to be the main cause for the low systemic availability. It is a right candidate to formulate into a SMEDDS formulation.
- The aim of the present study was to formulate, optimize and characterize stable self microemulsifying drug delivery system (SMEDDS) in order to enhance solubility as well as dissolution rate of this highly lipophilic drug, using single non-ionic surfactant with the use of a co-surfactant.

Future Plan

- ✓ Scale up studies of the optimized formulation
- ✓ *In vivo* studies and *in vitro – in vivo* correlation studies.
- ✓ To study the Bio equivalence study to optimize further.

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