Chapter 1

Introduction

Page no. 1 to 42
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Introduction

1.1. INTRODUCTION TO DRUG DELIVERY SYSTEM

In recent years, drug discovery program has dramatically undergone changes from “empirical-based” to “knowledge-based” rational drug design. Advances in biotechnology and combinatorial synthetic approaches, clubbed with high throughput screening (HTS) for pharmacological activity, have resulted in increasing number of diverse new chemical entities (NCEs). However, this rational design of molecules does not necessarily mean rational drug delivery, since the drug molecules do not always deliver themselves (Davis and Illum, 1998). Drug development in the past used to be initiated after identification of most active molecule. However this approach lead to a number of drawbacks with the problems being that many molecules which are put into development had poor physicochemical (solubility, stability) and biopharmaceutical (permeability and enzymatic stability) properties, as a consequence of which about 40% of NCEs fail to reach the market place (Prentis et al., 1988). Many investigational new drugs (INDs) fail during preclinical and clinical development, with an estimated 46% of compounds entering clinical development are dropped due to unacceptable efficacy and 40% due to safety reasons (Kennedy 1997). Oral delivery of such drugs is also frequently associated with low bioavailability, high intra- and inter-subject variability, and a lack of dose proportionality (Kommuru et al., 2001).

Currently a number of technologies are available to deal with the poor solubility, dissolution rate and bioavailability of insoluble drugs. Various formulation strategies reported in the literature include, incorporation of a drug in oils (Burcham et al., 1997), solid dispersions (Serajuddin et al., 1988), emulsions (Myers and Stella 1992b), liposomes (Schwendener and Schott 1996), use of cyclodextrins (Perng et al., 1998), coprecipitates (Nazzal et al., 2002), micronization (McInnes et al., 1982; Vogt et al., 2008), nanoparticles (Shabouri 2002), permeation enhancers (Aungst 2000) and lipid-based vehicles (Chakrabarti and Belpaire, 1978; Tokumura et al., 1987; Trull et al., 1995).

1.2. LIPID-BASED DRUG DELIVERY SYSTEM

Lipid-based drug delivery systems are experiencing a resurgence of interest lately (Hauss, 2007; Humberstone and Charman, 1997; Kreilgaard, 2002; Lawrence and Rees, 2000;

However, much attention has been focused on lipid-based formulations, with particular emphasis on self-emulsifying drug delivery systems or SEDDS, which were shown to improve the oral bioavailability of many drugs, viz. halofantrine (Khoo et al., 1998), ontazolest (Hauss et al., 1998), cyclosporine (Klauser et al., 1997), and progesterone (MacGregor et al., 1997).

Lipid formulations are a diverse group of formulations with a wide variety of properties and usually consist of a mixture of excipients, ranging from triglyceride oils through mixed glycerides, lipophilic surfactants, hydrophilic surfactants and co-solvents (Pouton, 2000). Lipid-based formulations can decrease the intrinsic limitations of slow and incomplete dissolution of poorly water soluble drugs by facilitating the formation of solubilised phases from which absorption takes place. The achievement of such phases will not essentially take place from the formulation itself, but alternatively from taking the advantage of the intra luminal processing to which lipids are subjected (Humberstone and Charman, 1997). The extent of drug absorption from lipid vehicles is significantly affected by the dispersibility of the administered lipid and drug. On the other hand, because of the inherent physical instability, the large volume of the two phase emulsion, and the poor precision of dose, the use of conventional emulsions is problematic. A formulation approach for avoiding such restrictive problems is the use of microemulsions or self-emulsifying drug delivery systems (SEDDS). The most famous example of a microemulsion based system is the Neoral® formulation of Cyclosporine, which result in replacement of Sandimmune® (Choc, 1997; Postolache et al., 2002).

1.3. CLASSIFICATION OF LIPID-BASED DRUG DELIVERY SYSTEM
Lipid system includes triglycerides, mono and diglycerides, lipophilic surfactants, hydrophilic surfactants and co-solvents; excipients with a wide variety of physicochemical properties. A classification system was introduced in 2000 to help identify the critical performance characteristics of lipid systems (Pouton, 2000). Table 1.1 is an updated version of what could reasonably be called the lipid formulation classification system (LFCS). Briefly, Type I formulations are oils which require to be digested, Type II formulations are water-insoluble self-emulsifying drug delivery systems (SEDDS), Type
III systems are SEDDS or self-microemulsifying drug delivery systems (SMEDDS) which contain some water-soluble surfactants and/or co-solvents (Type IIIA) or a greater proportion of water soluble components (Type IIIB). Table 1.1 includes an additional category (Type IV) to represent the recent trend towards formulations which contain predominantly hydrophilic surfactants and co-solvents. Type IV formulations contain no oils and represent the most extremely hydrophilic formulations. The advantage of blending a surfactant with a co-solvent to give a Type IV formulation is that the surfactant offers much greater good solvent capacity on dilution (as a micellar solution) than the co-solvent alone. The co-solvent is useful to facilitate dispersion of the surfactant, which is likely to reduce variability and irritancy caused by high local concentrations of surfactant. A Type IV formulation is useful for drugs which are hydrophobic but not lipophilic, though it is necessary to bear in mind that Type IV formulations may not be well-tolerated if the drug is to be used on a chronic basis. An example of a Type IV formulation is the current capsule formulation of the HIV protease inhibitor amprenavir (Agenerase, GSK) (Strickley, 2004). For this clinical indication the benefit clearly outweighs the risk. The general characteristics, advantages and disadvantages of each type of lipid formulation are shown in Table 1.2.

The performance of lipid formulations, and the fate of the drug in the gastrointestinal tract, depends on the physical changes that occur on dispersion and dilution of the formulation, and the influence of digestion on drug solubilisation. The main advantage of lipid formulation is that the drug can remains in solution form throughout its period in the gastrointestinal tract. However, if precipitation occurs at any stage the advantage of a lipid formulation is lost. Precipitation of drug is more prevalent from lipid systems which contain more hydrophilic excipients. Therefore, utmost care is needed with lipid formulation because such excipients are often used to improve the solvent capacity of the formulation, to increase the dose that can be administered in a single capsule.
Table 1.1: The proposed lipid formulation classification system (LFCS) showing typical composition of various types of lipid formulations

<table>
<thead>
<tr>
<th>Excipient composition (%)</th>
<th>Type I</th>
<th>Type II</th>
<th>Type IIIA</th>
<th>Type IIIB</th>
<th>Type IV</th>
</tr>
</thead>
<tbody>
<tr>
<td>Oils: triglycerides or mixed mono and diglycerides</td>
<td>100</td>
<td>40-80</td>
<td>40-80</td>
<td>&lt; 20</td>
<td>-</td>
</tr>
<tr>
<td>Water-insoluble surfactants (HLB ≤ 12)</td>
<td>-</td>
<td>20-60</td>
<td>-</td>
<td>-</td>
<td>0-20</td>
</tr>
<tr>
<td>Water-soluble surfactants (HLB &gt; 12)</td>
<td>-</td>
<td>-</td>
<td>20-40</td>
<td>20-50</td>
<td>30-80</td>
</tr>
<tr>
<td>Hydrophilic cosolvents (e.g. PEG, propylene glycol, transcutol)</td>
<td>-</td>
<td>-</td>
<td>0-40</td>
<td>20-50</td>
<td>0-50</td>
</tr>
<tr>
<td>Particle size of dispersion</td>
<td>Coarse</td>
<td>100-250</td>
<td>100-250</td>
<td>50-100</td>
<td></td>
</tr>
<tr>
<td>Significance of aqueous dilution</td>
<td>Limited importance</td>
<td>Solvent capacity unaffected</td>
<td>Some loss of solvent capacity</td>
<td>Significant phase changes and potential loss of solvent capacity</td>
<td>Micellar solution</td>
</tr>
<tr>
<td>Significance of digestibility</td>
<td>Crucial requirement</td>
<td>Not crucial but likely to occur</td>
<td>Not crucial but may be inhibited</td>
<td>Not required and not likely to occur</td>
<td>Limited digestion</td>
</tr>
</tbody>
</table>

Table 1.2: Characteristic features, advantages and disadvantages of the various types of ‘lipid’ formulations

<table>
<thead>
<tr>
<th>LFCS type</th>
<th>Characteristics</th>
<th>Advantages</th>
<th>Disadvantages</th>
</tr>
</thead>
<tbody>
<tr>
<td>Type I</td>
<td>Non-dispersing, requires digestion</td>
<td>GRAS status; simple; excellent capsule compatibility</td>
<td>Formulation has poor solvent capacity unless drug is highly lipophilic</td>
</tr>
<tr>
<td>Type II</td>
<td>SEDDS without water-soluble components</td>
<td>Unlikely to lose solvent capacity on dispersion</td>
<td>Turbid o/w dispersion (0.25-2 µm particle size)</td>
</tr>
<tr>
<td>Type IIIA</td>
<td>SEDDS/SMEDDS with water-soluble components</td>
<td>Clear or almost clear dispersion; drug absorption without digestion</td>
<td>Possible loss of solvent capacity on dispersion; less easily digested</td>
</tr>
<tr>
<td>Type IIIB</td>
<td>SMEDDS with water-soluble components and low oil content</td>
<td>Clear dispersion; drug absorption without digestion</td>
<td>Likely loss of solvent capacity on dispersion</td>
</tr>
<tr>
<td>Type IV</td>
<td>Oil-free formulation based on surfactants and co-solvents</td>
<td>Good solvent capacity for many drugs; disperses to micellar solution</td>
<td>Loss of solvent capacity on dispersion; may not be digestible</td>
</tr>
</tbody>
</table>

1.4. DIGESTION AND SOLUBILISATION OF LIPID-BASED FORMULATIONS IN THE SMALL INTESTINE

The solubilization capacity of the digestive system is considerable and its presence has an effect on the dissolution and absorption of lipophilic drugs from all formulations (Embleton and Pouton, 1997). The solubilising power is greater after a fatty meal, hence, food often has a positive effect on bioavailability of dissolution rate-limited BCS class II drugs. Bile salt concentrations in the gut are 3–5 mM on a fasted stomach and approximately 15 mM after food. Formulae for simulation of intestinal fluids in a fasted subject (FaSSIF) and fed subject (FeSSIF) have been used as alternative dissolution media (Dressman and Reppas, 2000), indicating that the dissolution of lipophilic drugs from conventional tablets is correlated with the availability of bile salt micelles (for example
Figure 1.1). A well-designed amorphous or lipid formulation presents the drug as a molecular dispersion, so the corresponding issue is whether the drug can be transferred to the mixed micellar system as the formulation is diluted into the aqueous phase. The surfactant components would be expected to interact with mixed bile salt micelles, which may result in a change in its structure and solubilization capacity (Lim and Lawrence, 2004). There are a few reports which shed light on the structures that result from interaction of non-ionic surfactants with mixed micelles.

**Figure 1.1:** Dissolution profile of Romazin tablets (troglitazone 200 mg) (Nicolaides et al., 1999). An example of how the rate and extent of dissolution from solid dosage forms is influenced by the likely components in the gut lumen. The presence of bile salt micelles in FaSSIF and FeSSIF increase both rate and extent. The higher concentrations of bile salt micelles in FeSSIF, representing the fed intestine, have a profound effect.

### 1.5. SELF EMULSIFYING DRUG DELIVERY SYSTEM

Self-emulsifying drug delivery systems (SEDDS) are mixtures of oils and surfactants, ideally isotropic, and sometimes containing co-solvents, which emulsify spontaneously to produce fine oil-in-water emulsions when introduced into aqueous phase under gentle agitation (Gershanik and Benita 2000; Gursoy and Benita, 2004; Shah et al., 1994; Craig et al., 1993). Recently, SEDDS have been formulated using medium chain triglyceride oils and non-ionic surfactants, the latter being less toxic. Upon peroral administration, these systems form fine emulsions (or micro-emulsions) in gastro-intestinal tract (GIT) with mild agitation provided by gastric mobility (Shah et al., 1994). Fine oil droplets would pass rapidly from the stomach and promote wide distribution of the drug throughout the GI tract, thereby minimizing the irritation frequently encountered during extended contact.
between bulk drug substances and the gut wall. The basic difference between self emulsifying drug delivery systems (SEDDS) also called as self emulsifying oil formulation (SEOF) and SMEDDS is SEDDS typically produce opaque emulsions with a droplet size between 100 and 300 nm while SMEDDS form transparent micro emulsions with a droplet size of less than 50 nm also the concentration of oil in SMEDDS is less than 20 % as compared to 40-80% in SEDDS. Potential advantages of these systems include enhanced oral bioavailability enabling reduction in dose, more consistent temporal profiles of drug absorption, selective targeting of drug(s) toward specific absorption window in GIT, and protection of drug(s) from the hostile environment in gut (Patil et al., 2004; Pouton and Charman, 1997).

Advantages
1) SMEDDS is a novel approach to improve water solubility and ultimate bioavailability of drugs for which water is a rate-limiting step. They have the ability to present the drug to GIT in 1-100 nm globule size and the subsequent increase in specific area enables more efficient drug transport through the intestinal aqueous boundary layer leading to improvement in bioavailability.

2) Many drugs show large inter-subject and intra-subject variation in absorption leading to fluctuation in plasma profile. Food is a major factor affecting therapeutic performance of the drug in the body. SMEDDS produce reproducible plasma profile.

3) Fine oil droplets empty rapidly from the stomach and promote wide distribution of the drug through the intestinal tract and thereby minimizing irritation frequently encountered with extended contact of drugs with gut wall.

4) Ease of manufacture and scale up is one of the most important advantages that make SMEDDS unique, when compared to other drug delivery system like solid dispersion, liposomes, nanoparticles etc; dealing with improved bioavailability. SMEDDS require very simple and economical manufacturing facility like mixture with agitator and volumetric liquid filing equipment for large-scale manufacturing. This is of interest to pharmaceutical industry.

5) SMEDDS has potential to deliver peptides that are prone to enzymatic hydrolysis in GIT.

6) When polymer is incorporated in the composition of SMEDDS, it gives prolonged release of medicaments.

7) Enhanced oral bioavailability results in dose reduction.
8) Selective targeting of drugs towards specific absorption window in GIT.

**Limitations**

One of the obstacles for the development of self emulsifying drug delivery systems (SEDDS) and other lipid-based formulations is the lack of good predicative *in vitro* models for assessment of the formulations (Tang et al., 2008; Zhang et al., 2008). Traditional dissolution methods do not work, because these formulations potentially are dependent on digestion in the gut, prior to release of the drug. To mimic this, an *in vitro* model simulating the digestive processes of the duodenum has been developed. This *in vitro* model needs further refinement and validation before its strength can be evaluated. Further development will be based on *in vitro - in vivo* correlations and therefore different prototype lipid based formulations need to be developed and tested *in vivo* in a suitable animal model (Patil et al., 2007; Tang et al., 2007).

Few other drawbacks are chemical instabilities of drugs and high surfactant concentrations in formulations (approximately 30-60%) which irritate GIT. Moreover, volatile co-solvents in the conventional self-microemulsifying formulations are known to migrate into the shells of soft or hard gelatin capsules, resulting in the precipitation of the lipophilic drugs. The precipitation tendency of the drug on dilution may be higher due to the dilution effect of the hydrophilic solvent. At the same time, formulations containing several components become more challenging to validate.

**Mechanism of self-emulsification**

The process by which self-emulsification takes place is not well understood. Self-emulsification occurs when the entropy change that favors dispersion is greater than the energy required to increase the surface area of the dispersion. In addition, the free energy of a conventional emulsion formation is a direct function of the energy required to create a new surface between the two phases and can be described by equation as given below (Reiss, 1975).

\[
\Delta G = \sum_{i} N_i \pi r_i^2 \sigma
\]

Where \( \Delta G \) is the free energy associated with the process (ignoring the free energy of mixing), \( N \) is the number of droplets of radius, \( r \), and \( \sigma \) represents the interfacial energy.
With time, the two phases of the emulsion will tend to separate, in order to reduce the interfacial area, and also, the free energy of the systems. Therefore, the emulsions resulting from aqueous dilution are stabilized by conventional emulsifying agents, which form a monolayer around the emulsion droplets, and hence, reduce the interfacial energy, as well as providing a barrier to coalescence. In the case of self-emulsifying systems, the free energy required to form the emulsion is either very low and positive, or negative (then, the emulsification process occurs spontaneously) (Craig et al., 1995). Emulsification requiring very little input energy involves destabilization through contraction of local interfacial regions. For emulsification to occur, it is necessary for the interfacial structure to have no resistance to surface shearing (Dabros et al., 1999).

It has been suggested that the ease of emulsification could be associated with the ease by which water penetrates into the various liquid crystalline (LC) or gel phases formed on the surface of the droplet (Rang and Miller, 1999). According to Wakerly et al (1986), the addition of a binary mixture (oil/non-ionic surfactant) to water results in interface formation between the oil and aqueous-continuous phases, followed by the solubilization of water within the oil phase owing to aqueous penetration through the interface. This will occur until the solubilization limit is reached close to the interface. Further aqueous penetration will result in the formation of the dispersed LC phase. As the aqueous penetration proceeds, eventually all material close to the interface will be LC, the actual amount depending on the surfactant concentration in the binary mixture. Once formed, rapid penetration of water into the aqueous cores, aided by the gentle agitation of the self-emulsification process, causes interface disruption and droplet formation. The high stability of these self-emulsified systems to coalescence is considered to be due to the LC interface surrounding the oil droplets. The involvement of the LC phase in the emulsion formation process was extensively studied by Pouton et al. (Rang and Miller, 1999; Wakerly et al., 1986). Later, Craig et al. used the combination of particle size analysis and low frequency dielectric spectroscopy (LFDS) to examine the self-emulsifying properties of a series of Imwitor 742 (a mixture of mono- and diglycerides of capric and caprylic acids)/Tween 80 systems (Craig et al., 1993; Craig et al., 1995). The dielectric studies have provided evidence that the formation of the emulsions may be associated with LC formation, although the relationship was clearly complex (Craig et al., 1995).
1.6. COMPONENTS OF SEDDS

Natural product oils

A number of natural product oils, derived primarily from plant sources and processed to remove impurities or to isolate various fractions of the original product, are available and suitable for use in encapsulated oral formulation products. Naturally occurring oils and fats are comprised of mixtures of triglycerides which contain fatty acids of varying chain lengths and degrees of unsaturation. The melting point of particular oil increases in proportion to the fatty acid chain length and decreases with increasing degree of unsaturation. However, unsaturation can make it susceptible to oxidation. Triglycerides are classified as short (< 5 carbons), medium (6–12 carbons), or long chain (> 12 carbons) and may be synthetically hydrogenated to decrease the degree of unsaturation, thereby conferring resistance to oxidative degradation. Separation of natural product oils into their component glyceride fractions is used to prepare excipients that maximize desirable physical and drug absorption-promoting properties (Greenberger et al., 1966) while minimizing susceptibility to oxidation.

Semi-synthetic lipid excipients

Several semi-synthetic liquid and thermo-softening (semisolid) excipients, most commonly prepared by chemically combining medium-chain saturated fatty acids or glycerides derived from natural product plant oils, with one or more hydrophilic chemical entities are currently available as pharmaceutical excipients for oral formulation development (Gibson, 2007). These excipients find application as drug-solubilizing vehicles, surfactants and wetting agents and as emulsifiers and co-emulsifiers in SEDDS and self-microemulsifying drug delivery systems (SMEDDS). They are generally well-suited for filling into both soft and hard gelatin or into HPMC capsules. Thermo-softening excipients, which melt in the range of 26–70 °C and exist as waxy semi-solids at ambient room temperatures, are typically filled into capsules in the molten state, with the excipient melting temperature limiting their use to hard gelatin capsules.

Fully-synthetic excipients

A number of fully-synthetic, monomeric and polymeric liquid and semi-solid excipients, most of which are glycolic in nature and relatively non-toxic, are used as solvents for formulating poorly water-soluble drugs. These excipients can be used alone or in
combination with other lipid excipients to improve the overall solubilizing power of the formulation. However, their pronounced water miscibility can compromise formulation performance due to uncontrolled precipitation of the drug substance following dilution in the aqueous contents of the GIT. This results in dose-dependent bioavailability enhancement. A few examples of the most commonly applied excipients in this class and their applications follows. Among the polymeric glycol-based excipients finding pharmaceutical application, the polyethylene glycols (PEGs) are a versatile, well-characterized and widely applied class of solubilizers which are available as both liquids and thermo-softening semi-solids. The physical state of these excipients at ambient room temperature is determined by their molecular weights (Rowe and Sheskey, 2003a). PEGs ranging from 200 to 600 in molecular weight are liquid at ambient room temperature whereas those possessing molecular weights of 1000 or greater exist as thermo-softening semi-solids. In comparison to natural product oils, PEGs have the following disadvantages: they tend to be more chemically reactive; they can be more irritating to the GI mucosa than oils. PEGs are also known to contain varying levels of peroxide impurities and secondary products formed by auto-oxidation, which can contribute to chemical instability of the incorporated drug substance. These excipients are widely used in soft gelatin capsule formulations but find only limited use in conjunction with hard gelatin capsules due to their hygroscopicity and resultant effects on gelatin moisture content, which can compromise capsule physical integrity. Propylene glycol, a pharmaceutically-acceptable, monomeric solvent possessing humectant and plasticizing properties, finds application for soft gelatin capsule formulations of poorly water-soluble drugs (Rowe and Sheskey, 2003b). The poloxamers, which are co-polymers of polyoxyethylene and polyoxypropylene, possess both solvent and surfactant properties and thus find application in the oral delivery of poorly water-soluble drugs (Rowe and Sheskey, 2003c). As with the PEGs, they are available in a range of molecular weights which control the physical state of the excipient at room temperature. In addition to improving the bioavailability of poorly water-soluble drugs, they have found application in modified release formulations (Anderson et al., 2001).

**Surfactants**

Non-ionic surfactants with a relatively high hydrophilic - lipophilic balance (HLB) have been suggested for the design of self-dispersing systems, where the various liquid or solid
ethoxylated polyglycolyzed glycerides and polyoxyethylene 20 oleate (Tween 80) are the most frequently used excipients (Table 1.3). Emulsifiers derived from natural sources are expected to be safer than synthetic ones and are recommended for self dispersing liquid formulation (SDLF) use, despite their limited ability to self-emulsify (Hauss et al., 1998; Yuasa et al., 1994). Non-ionic surfactants are known to be less toxic compared to ionic surface-active agents, but they may cause moderate reversible changes in intestinal wall permeability (Swenson et al., 1994; Wakerly et al., 1986). Amemiya et al. have proposed a new vehicle based on a fine emulsion using minimal surfactant content (3%) to avoid the potential toxicological problems associated with high surfactant concentration (Amemiya et al., 1998). The usual surfactant concentration in self-emulsifying formulations required to form and maintain an emulsion state in the GI tract ranged from 30 to 60% w/w of the formulation. A large quantity of surfactant may irritate the GI tract. Thus, the safety aspect of the surfactant vehicle should be carefully considered in each case. The high HLB and subsequent hydrophilicity of surfactants is necessary for the immediate formation of o/w droplets and/or rapid spreading of the formulation in the aqueous environment, providing a good dispersing/self-emulsifying performance. The surface-active agents are amphiphilic by nature, and they are therefore usually able to dissolve and even solubilize relatively high quantities of the hydrophobic drugs. The latter is of prime importance for preventing precipitation within the GI lumen and for the prolonged existence of the drug molecules in the soluble form, vital for effective absorption (Serajuddin et al., 1988; Shah et al., 1994). The lipid mixtures with higher surfactant and co-surfactant/oil ratios lead to the formation of self-microemulsifying formulations (SMEDDS) (Constantinides, 1995). Formulations consisting only of the surfactant mixture may form emulsions or microemulsions (when surfactants exhibit different low and high HLB) (Farah et al., 1994), micelle solution (Aungst et al., 1994) or, in some particular cases, niosomes, which are non-ionic, surfactant-based bilayer vehicles (Uchegbu and Vyas, 1998).

Co-solvents
Relatively high surfactant concentrations (usually more than 30% w/w) are required to produce an effective self-emulsifying system (Gershanik and Benita, 2000). Organic solvents, suitable for oral administration (ethanol, propylene glycol (PG), polyethylene glycol (PEG), etc.) may help to dissolve large amounts of either the hydrophilic surfactant or the drug in the lipid base. These solvents sometimes play the role of the co-surfactant in
the microemulsion systems, although alcohol-free self-emulsifying microemulsions have also been described in the literature (Constantinides, 1995). Such systems may exhibit some advantages over the previous formulations when incorporated in capsule dosage forms, since alcohol and other volatile co-solvents comprised in the conventional self-emulsifying formulations are known to migrate into the shells of soft gelatin, or hard, sealed gelatin capsules, resulting in the precipitation of the lipophilic drug. On the other hand, the lipophilic drug dissolution ability of the alcohol free formulation may be limited.
Table 1.3: Composition of SDLFs that led to oral bioavailability enhancement of lipophilic drugs in *in-vivo* models

<table>
<thead>
<tr>
<th>Delivery system</th>
<th>Oil</th>
<th>Surfactant(s)</th>
<th>% w/w Solvent(s)</th>
<th>Model Drug</th>
<th>Drug content (%)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>SEDDS</td>
<td>Mixture of mono- and di-glycerides of oleic acid</td>
<td>Solid, polyglycolized mono-, di- and triglycerides (HLB = 14)</td>
<td>80 or 20</td>
<td>Ontazolast</td>
<td>7.5</td>
<td>Hauss et al., 1998</td>
</tr>
<tr>
<td>Surfactant/Solvent dispersion</td>
<td>-</td>
<td>Tween 80 (HLB = 15)</td>
<td>NA</td>
<td>Ethanol PG</td>
<td>Retinol (Vitamin A), riboflavin (Vitamin B2)</td>
<td>NA</td>
</tr>
<tr>
<td>Surfactant dispersion</td>
<td>-</td>
<td>Solid, polyglycolized mono-, di- and triglycerides (HLB = 14)</td>
<td>81</td>
<td>A-pentyl-3-(2-quinolinyl-methoxy)-benzenemethanol</td>
<td>19</td>
<td>Serajuddin et al., 1988</td>
</tr>
<tr>
<td>Sandimmune® (SEDDS)</td>
<td>Olive oil</td>
<td>Polyglycolized glycerides (HLB = 3/4)</td>
<td>30</td>
<td>Ethanol</td>
<td>Cyclosporin A</td>
<td>10</td>
</tr>
<tr>
<td>Lipid dispersion</td>
<td>Medium chain monacyl glycerol</td>
<td>Soybean phosphatidyl choline</td>
<td>30</td>
<td>-</td>
<td>Hexarelin</td>
<td>11.8</td>
</tr>
<tr>
<td>SEDDS</td>
<td>Medium chain saturated fatty acids, peanut</td>
<td>Medium chain mono- and di-glycerides, Tween 80, PEG-25 glyceryl trioleate, Polyglycolyzed glycerides (HLB = 6-14)</td>
<td>5-60</td>
<td>Ro 15-0778, a naphthalene derivative</td>
<td>5</td>
<td>Shah et al., 1994</td>
</tr>
<tr>
<td>SEDDS</td>
<td>Medium chain fatty acids</td>
<td>PEG-25 glyceryl trioleate</td>
<td>25</td>
<td>WIN 54954 (5-(5-(2,6-dichloro-4-(dihydro-2-oxazolyl)phentoxypentyl)-3-methyisoxazole</td>
<td>35</td>
<td>Charman et al., 1992</td>
</tr>
<tr>
<td>SMEDDS</td>
<td>-</td>
<td>Polyglycolyzed glycerides (HLB = 1-14)</td>
<td>96</td>
<td>-</td>
<td>Indomethacin</td>
<td>4</td>
</tr>
<tr>
<td>Surfactant dispersion</td>
<td>-</td>
<td>Polyglycolyzed glycerides (HLB = 14)</td>
<td>79.5</td>
<td>DMP 323 (HIV protease inhibitor)</td>
<td>7.5 or 12.5</td>
<td>Aungst et al., 1994</td>
</tr>
<tr>
<td>Neoral® formulation (SMEDDS)</td>
<td>Hydrolyzed corn oil</td>
<td>Polyglycolyzed glycerides, POE-castor oil derivative</td>
<td>NA</td>
<td>Glycerol</td>
<td>Cyclosporin A</td>
<td>10</td>
</tr>
<tr>
<td>Neoral® formulation (SMEDDS)</td>
<td>Hydrolyzed corn oil</td>
<td>Polyglycolyzed glycerides, POE-castor oil derivative</td>
<td>NA</td>
<td>Ethanol</td>
<td>Cyclosporin A</td>
<td>10</td>
</tr>
<tr>
<td>Positively charged SEDDS</td>
<td>Ethyl oleate</td>
<td>Tween 80</td>
<td>25</td>
<td>Ethanol</td>
<td>Cyclosporin A</td>
<td>10</td>
</tr>
<tr>
<td>Positively charged SEDDS</td>
<td>Ethyl oleate</td>
<td>Tween 80</td>
<td>25</td>
<td>Ethanol</td>
<td>Progesterone</td>
<td>2.5</td>
</tr>
</tbody>
</table>
1.7. BIOPHARMACEUTICAL ISSUES AND CHOICE OF A FORMULATION

_Dose of drug_

The optimum formulation for each drug will depend on a number of factors; on the required dose, on which types of formulation have sufficient solvent capacity to allow for formulation of a unit dose, and in particular on the fate of the drug after these formulations have been administered to the gut. In many instances the choice of formulation will be limited by solvent capacity, and in others the drug will not be sufficiently soluble in any lipid formulations. Generally the most difficult drugs are those which have limited solubility in both water and lipid (typically with log $P$ values of approximately 2). It is unlikely that lipid formulation will be of value for such drugs. In contrast, more hydrophobic drugs may permeate lipid bilayers freely but dissolve very slowly in the lumen of the gut. It has been common in the past for formulators to be presented with the challenge of formulating a high dose oral product for a new hydrophobic drug, in circumstances when the dose has been estimated using the results of early pre-clinical animal studies. Such studies may have been conducted with an inadequate formulation, such as a crude aqueous suspension, from which bioavailability is poor. In these circumstances it is wise to anticipate that bioavailability may be much greater from a lipid system, which may allow pharmacological activity to be achieved with a lower dose. This is particularly important for drugs with high log $P$, when in the first instance the solvent capacity of lipid formulations appear to fall short of the required dose.

If the drug is potent and is sufficiently soluble in Type I, Type II or Type III systems then a choice will need to be made between these options. The most important consideration should be to avoid the precipitation of a drug, but secondary consideration is whether or not rapid absorption is desirable. Typically Type II or Type III systems will undergo gastric emptying earlier and will be in a colloidal state earlier than Type I systems. An emulsifying system is likely to result in more rapid absorption and higher peak concentrations of drug. If the drug has a low therapeutic index this may be undesirable, which argues in favour of a Type I formulation.

_Significance of droplet size_

Tarr and Yalkowsky (1989) were able to demonstrate in a gut perfusion experiment that emulsion droplet size affected the rate of absorption of cyclosporine A. Absorption was
more rapid from the finer of two emulsions, though the emulsions compared were both relatively coarse. The situation is very different when a small volume of a lipid formulation is administered in a capsule. The contents of the capsule will be emptied into the digestive environment of the upper small intestine, so that the most important factor may not be the size of the particles in the initial dispersion, but rather their susceptibility to digestion and/or solubilisation by mixed micelles of bile salts and phospholipids. One consequence of reducing the oil content and including surfactants and co-solvents is that the droplets become less susceptible to digestion. This means that self-emulsifying systems are dependent on the initial emulsification process to produce a colloidal dispersion. It is assumed that the droplet size should be as fine as possible, and there is some evidence that this assumption holds in the case of cyclosporine A. The drug was more available from the ‘Neoral’ formulation than the earlier ‘Sandimmune’ formulation, which was a coarsely emulsifying system (Mueller et al., 1994). One possible explanation for this observation may be that the coarse emulsion produced by the ‘Sandimmune$^\text{TM}$’ formulation could not be reduced to colloidal dimensions, due to limited digestion. Type IIIB formulations generally produce the finest dispersions, due to their high content of water-soluble solubilising agents (Constantinides, 1995).

The risk of precipitation

Consider the example of a hydrophobic drug dissolved in a pure co-solvent such as polyethylene glycol or propylene glycol. When the formulation is added to water, the solvent capacity of the mixture falls, approximately, logarithmically as the formulation is diluted into water. The result is precipitation of the drug. It is much more difficult to predict the fate of the drug on dispersion of a Type IIIA lipid formulation; perhaps consisting of a drug dissolved in 30% medium chain triglycerides, 40% mixed partial glycerides, and 30% hydrophilic surfactant. The hydrophilic surfactant will be substantially separated from the oily components, forming a micellar solution in the continuous phase. Does this lower the overall solvent capacity for the drug? That will depend on the log $P$ of the drug, and to what extent the surfactant was contributing to its solubilisation within the formulation.
Role of lipolysis and solubilisation in bile

Digestion of dietary triglycerides in the small intestine is very rapid, and many other non-ionic esters, such as mixed glycerides and surfactants will be substrates for pancreatic lipase (Embleton and Pouton, 1997). Digestion of formulations will inevitably have a profound effect on the state of dispersion of the lipid formulation, and the fate of the drug (MacGregor et al., 1997). The natural process of digestion offers the possibility that very hydrophobic drugs could be taken up into the lymphatic system by partitioning into chylomicrons in the mesentery (Porter and Charman, 1997). This is expected to be a mechanism of absorption for drugs with log $P$ values greater than 6, and has been demonstrated for the anti-malarial compound halofantrine (Porter et al., 1996).

However, it is possible that digestion of a lipid formulation could reduce the solubility of the drug in the gut lumen, which would result in precipitation of the drug and a decrease in the absorption rate. More work needs to be carried out to establish which drugs are precipitated on digestion. For such compounds Type II or Type III systems might be preferable, since the presence of surfactants can inhibit digestion of the oil within the formulation (MacGregor et al., 1997). Type III systems such as ‘Neoral’ have been shown to act independently of bile, which suggests that they are not necessarily digested before the drug is absorbed (Trull et al., 1995).

Drug absorption from SDLFs

Numerous bioavailability studies carried out in animals suggested that hydrophobic drugs are better absorbed when administered as o/w emulsions (Hauss et al., 1994; Myers and Stella, 1992b; Palin et al., 1986; Stella et al., 1978; Toguchi et al., 1990a; Toguchi et al., 1990b). The improvement of the plasma profile reproducibility of WIN 54954 following administration in SEDDS compared to PEG solution was emphasized by Charman et al. (Charman et al., 1992), while others reported a higher bioavailability of the hydrophobic drugs after administration as SEDDS (Fischl et al., 1997; Hauss et al., 1998; Shah et al., 1994). In in-vivo absorption studies in non-fasting dogs for a lipophilic drug, RO15-0778, a naphthalene derivative (Shah et al., 1994), SEDDS gave at least a three-fold greater $C_{\text{max}}$ and AUC than the drug in any other liquid or solid oral dosage form (Table 1.4) (Shah et al., 1994). The absorption in rats of ontazolast, a poorly water-soluble, lipophilic anti-inflammatory compound (a potent calcium ionophore A23187-stimulated leukotriene B4
inhibitor), as a function of lipid-based delivery system composition was investigated by Hauss et al (1998). The bioavailability of this drug was significantly enhanced by all lipid-based formulations: the emulsion, glyceryl oleate (Peceol) solution and both semisolid SEDDS, containing either 20/80 or 50/50 Peceol/Gelucire 44/14 (PEG-32 glyceryl laurate) compared to suspension formulation. The absorption of the poorly water-soluble experimental drugs REV 5901 (a-pentyl-3-(2-quinolinylmethoxy)-benzenemethanol) and DMP 323 (HIV protease inhibitor) in dogs from capsules containing solid dispersion with Gelucire 44/14 was higher than that from PEG-based formulations (Serajuddin et al., 1988). In multiple dosage studies carried out in HIV-infected patients, the administration of SEDDS formulation resulted in a larger AUC, and a higher \( C_{\text{min}} \) and \( C_{\text{max}} \), of the HIV protease inhibitor SC-52151 (a urea-based peptide-mimetic compound) than the corresponding elixir formulation (Fischl et al., 1997). A recently designed freeze-dried formulation, consisting of olive oil and egg albumin, was shown to markedly enhance the oral bioavailability of several hydrophobic drugs (Tsuji et al., 1996).

### Table 1.4: Pharmacokinetic parameters of a lipophilic naphthalene derivative (Ro 15 0778) from different formulations in non-fasting dogs

<table>
<thead>
<tr>
<th>Formulation</th>
<th>( C_{\text{max}} ) (mg/mL)</th>
<th>( T_{\text{max}} ) (h)</th>
<th>( AUC ) (µg.h/mL)</th>
<th>Relative bioavailability (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>SEDDS</td>
<td>5.57</td>
<td>2.50</td>
<td>29.77</td>
<td>389.0</td>
</tr>
<tr>
<td>Drug solution in PEG 400 (Control)</td>
<td>1.44</td>
<td>2.00</td>
<td>7.64</td>
<td>100.0</td>
</tr>
<tr>
<td>Capsule formulation of wet-milled spray dried powder</td>
<td>0.78</td>
<td>3.00</td>
<td>2.69</td>
<td>35.3</td>
</tr>
<tr>
<td>Tablet of micronized drug</td>
<td>0.58</td>
<td>2.00</td>
<td>1.32</td>
<td>17.2</td>
</tr>
</tbody>
</table>

**Influence of the lipid vehicle**

Effect of the oil vehicle on lipophilic drug pharmacokinetic parameters may be discussed with respect to the effect of lipid composition and the efficiency of the carrier system. The effect of lipids on the bioavailability of orally administered drugs is highly complex due to numerous mechanisms by which the lipids can alter the biopharmaceutical characteristics of the drug. They include a decreased rate of gastric emptying, an increased dissolution rate of the drug and solubility in the intestinal fluid, and the formation of lipoproteins promoting the lymphatic transport of highly lipophilic drugs (Hauss et al., 1998; Hauss et al., 1994).
Factors such as the acid chain length of a triglyceride, the saturation degree and the volume of lipid administered may affect the drug absorption profile and its blood/lymph distribution. Three major processes can occur in the intestine: large fat droplets are further emulsified by bile salts, monoglycerides, cholesterol, lecithin and lysolecithin to produce droplets with a mean diameter of 0.5-1 mm. These droplets are then metabolized by pancreatic lipase, an enzyme with a molecular weight of about 40 000 Da (Maio and Carrier, 2011). The dispersed oil droplet fragments form mixed micelles with bile salts (Senior, 1964).

*Lymphatic absorption pathway*

Charman et al (1992 and 1986) proposed that drug candidates for lymphatic transport should have a log $P > 5$ and, in addition, a triglyceride solubility $> 50$ mg/ml. The importance of lipid solubility was illustrated by a comparing the lymphatic transport of dichloro diphenyl trichloroethane (DDT, log $P$ 6.19) with hexachlorobenzene (HCB, log $P$ 6.53). While both compounds have similar log $P$ values, the difference in lymphatic transport on administration in oleic acid, 33.5% of the dose in the case of DDT and 2.3% with HCB, was attributed to the 13-fold difference in triglyceride solubility (Charman et al., 1986). However, combination of a high log $P$ and high triglyceride solubility does not always guarantee significant lymphatic transport. Penclomedine, an experimental cytotoxic agent with a log $P$ of 5.48 and a triglyceride solubility of 175 mg/ ml, was poorly transported in the intestinal lymph, ~3% of the dose (Myers and Stella, 1992a). Khoo *et al.* showed significant lymphatic transport of the poorly lipid soluble (~1 mg/ml) HCl salt of halofantrine (Hf-HCl), following oral post-prandial administration to dogs. The authors suggest that the high level of lymphatic transport of Hf-HCl (43.7% of dose), which was similar to that of the lipid soluble Hf base, was due to conversion of Hf-HCl in the intestinal lumen, during lipolysis, to the more lipophilic free base, which then becomes associated with chylomicron production (Khoo *et al.*, 1999).

Partitioning of the absorbed drug into the lymph is hindered by the low rate of lymph fluid transport relative to that of blood, which is approximately 500-fold greater in the rat. Nevertheless, this way may be contributive for highly lipophilic drugs with a log $P$ of more than 5, and high triglyceride solubility (Hauss *et al.*, 1998; Hauss *et al.*, 1994; Toguchi *et al.*, 1990a). The extent of lymphatic absorption may be altered by the lipid
vehicle (Hauss et al., 1998; Hauss et al., 1994; Porter et al., 1996; Toguchi et al., 1990a). The rank order effect of the vehicles for the promotion of the lymphatic transport of halofantrine hydrochloride, a highly lipophilic anti-malarial agent, was micelles > emulsion > lipid solution (Porter et al., 1996). In another study, the soybean-based emulsion formulation produced significantly greater drug (ontazolast) transport into the lymph than Peceol/Tween 80 SEDDS formulations (Hauss et al., 1998). Evidently, the lipid composition of the vehicle, more so than the vehicle type, is the decisive factor for lymphatic transport promotion. It was shown that enhanced lymphatic transport is not necessarily reflected by proportionally elevated plasma AUC (Hauss et al., 1998; Hauss et al., 1994). The amount of ontazolast transported by the lymph with only Peceol solution was as high as that for the emulsion formulation, but the overall drug bioavailability from oil was lower than that with SEDDS or emulsion formulations. In the other study, the total quantity of CI-976 (2,2-dimethyl-N-(2,4,6-trimethoxyphenyl) dodecanamide), a poorly water-soluble lipid regulator, transported in the lymph and accumulated in the peripheral fat as a percentage of the dose administered, was much greater for the emulsion compared to the surfactant dispersion formulation, although the plasma AUC was higher for the dispersion formulation (Hauss et al., 1994).

Drug release
The release of a drug occurs following its partitioning to aqueous intestinal fluids during droplet transport and disintegration along the GI tract. Shah et al. (1994) proposed that the effective delivery of a drug from SEDDS is governed by two main factors: small particle size and the polarity of the resulting oil droplets, which permits a faster rate of drug release into the aqueous phase. The optimal polarity of the formulation is achieved by the appropriate combination of oil and surfactants. In o/w microemulsion formulations, the polarity of the oil phase is a less dominant factor, since the drug can reach the capillaries while still incorporated within the microemulsion droplets (Constantinides, 1995; Georgakopoulos et al., 1992; Vonderscher and Meinzer, 1994). Excess surfactant in a formulation may also promote the effective dispersion of drug in the lumen by the solubilisation process, in addition to drug spreading in oil droplets (Toguchi et al., 1990b). The solubilized drug does not precipitate in the lumen, and undergoes rapid absorption which is independent of the lipid digestion process. The presence of the lipid core in the dispersed system is not a prerequisite for achieving the best solubilization effect. In some
cases, therefore, the surfactant-based vehicle may be preferable, due to relative ease of formulation design compared to SEDDS/microemulsion formulations (Serajuddin et al., 1988). It was shown that the drug loading improvement of an o/w microemulsion over a micellar system appears to depend on the solubility of the drug in the dispersed oil phase, and is significant only for very lipophilic drugs (Malcolmson and Lawrence, 1993; Naylor et al., 1993).

**Positively charged SEDDS**

Multiple physiological studies have proved that the apical potential of absorptive cells, as well as that of all other cells in the body, is negatively charged with respect to the mucosal solution in the lumen (Aungst et al., 1988; Corbo et al., 1990). It was recently shown that positively charged emulsion droplets formed by appropriate SEDDS dilution undergo electrostatic interaction with the Caco-2 monolayer and the mucosal surface of the everted rat intestine (Gershanik et al., 1998; Gershanik et al., 2000). This formulation enhanced the oral bioavailability of progesterone in young female rats (Gershanik and Benita, 1996) and exhibited higher blood levels of Cyclosporine A (CsA) in perfused rats in comparison to the corresponding negatively charged formulation. Many aspects of these formulations are still unclear. Positively charged formulations differ from negatively charged formulations with respect to their interaction with biological components in the GI environment. Positively charged droplets should be attracted to the negatively charged physiological compounds naturally occurring in lumen (Gershanik et al., 1998). It was already shown by these authors that larger droplets (a few microns in size range) are less neutralized by mucin solutions of different concentrations than smaller droplets (submicron size range) formed by the same formulation (Gershanik et al., 1998).

**1.8. REGULATORY STATUS OF LIPID EXCIPIENTS**

Historically, excipients were considered inert substances that would be used mainly as diluents, fillers, binders, lubricants, coatings, solvents, and dyes, in the manufacture of drug products (Handbook of Pharmaceutical Excipients, 1986). Over the years, however, advances in pharmaceutical science and technology have facilitated the availability of a wide range of novel excipients. In some cases, known and/or unknown interactions can occur between an excipient and active ingredient, other inactive ingredient(s), biological surroundings, or even container closure system (Adkin et al., 1995; Aungst, 2000; Basit et
al., 2002; Bernkop-Schnurch and Kast, 2001; Chen et al., 2007; Hermeling et al., 2006; Hermeling et al., 2003; Martin-Facklam et al., 2002; Rege et al., 2001; Sharma et al., 2004; Tayrouz et al., 2003; Villalobos et al., 2005; Yu et al., 1999). Accordingly, it is now recognized that not all excipients are inert substances and some may be potential toxicants (http://www.fda.gov/cder/guidance/5544fnl.pdf.). The U.S. Food and Drug Administration (FDA) has published listings in the Code of Federal Regulations (CFR) for GRAS substances that are generally recognized as safe (USFDA, Title 21, Code of Federal Regulations, Part 182, 184, 186.). Over the years, the Agency also maintains a list entitled Inactive Ingredient Guide (IIG) for excipients that have been approved and incorporated in the marketed products (http://www.fda.gov/cder/drug/iig/default.htm; http://www.accessdata.fda.gov/scripts/cder/iig/index.cfm). This guide is helpful in that it provides the database of allowed excipients with the maximum dosage level by route of administration or dosage form for each excipient. Both GRAS listings and IIG information can be used by industry as an aid in developing drug products.

For new drug development purposes, once an inactive ingredient has appeared in an approved drug product for a particular route of administration, the inactive ingredient is not considered new and may require a less extensive review the next time it is included in a new drug product. For example, if a particular inactive ingredient has been approved in a certain dosage form with certain potency, a sponsor could consider it safe for use in a similar manner for a similar type of product. In general, nonclinical and clinical studies are required to demonstrate the safety of a new excipient before use. In this context, the U.S. FDA has recently published a guidance document for industry on the conduct of nonclinical studies for the safety evaluation of new pharmaceutical excipients (http://www.fda.gov/cder/guidance/5544fnl.pdf.). This guidance not only provides the types of toxicity data to be used in determining whether a potential new excipient is safe, but also describes the safety evaluations for excipients proposed for use in over-the-counter and generic drug products. The document also depicts testing strategies for pharmaceuticals proposed for short-term, intermediate, and long-term use. More importantly, this guidance highlights the importance of performing risk-benefit assessments on proposed new excipients in the drug products while establishing permissible and safe limits for the excipients. As illustrated, with proper planning, it is often possible to assess the toxicology of an excipient in a relatively efficient manner (http://www.fda.gov/cder/guidance/5544fnl.pdf.). Existing human data for some excipients
can substitute for certain nonclinical safety data. In addition, an excipient with documented prior human exposure under circumstances relevant to the proposed use may not require evaluation with a full battery of toxicology studies (http://www.fda.gov/cder/guidance/5544fnl.pdf.).

There is no process or mechanism currently in place within the FDA to independently evaluate the safety of an excipient. Instead, for a drug or biological product subject to pre-marketing approval, their excipients are reviewed and approved as ‘components’ of the drug or biological product in the application. From a scientific standpoint, the regulatory process is appropriate since excipients play an integral part to the formulation and cannot be reviewed separately from the drug product. This is particularly true for lipid excipients in view of their distinct physicochemical properties and potential complex interactions with other ingredients or physiological environment that may occur in vivo.

1.9. CURRENTLY MARKETED ORAL LIPID-BASED FORMULATION PRODUCTS

As determined in a recently published survey by Strickley, oral lipid-based formulations have been marketed for over 2 decades and currently comprise an estimated 2–4% of the commercially available drug products surveyed in 3 markets worldwide (Strickley, 2004 & 2007) (Table 1.5-1.7). These products accounted for approximately 2% (21 products total) of marketed drug products in the United Kingdom, 3% (27 products total) in the United States of America, and 4% (8 products total) in Japan. Strickley's survey revealed that the most frequently chosen excipients for preparing oral lipid-based formulations were dietary oils composed of medium- (e.g., coconut or palm seed oil) or long-chain triglycerides (e.g., corn, olive, peanut, rapeseed, sesame, or soybean oils, including hydrogenated soybean or vegetable oils), lipid soluble solvents (e.g., polyethylene glycol 400, ethanol, propylene glycol, glycerin), and various pharmaceutically-acceptable surfactants (e.g., Cremophor® EL, RH40, or RH60; polysorbate 20 or 80; D-α-tocopherol polyethylene glycol 1000 succinate (TPGS®); Span 20; various Labrafils®, Labrasol®, and Gelucires®). These formulations, which took the form of either bulk oral solutions or liquid-filled hard or soft gelatin capsules, were applied in instances where conventional approaches (i.e., solid wet or dry granulation, or water-miscible solution in a capsule) did not provide sufficient bioavailability, or in instances in which the drug substance itself was
an oil (e.g., dronabinol, ethyl icosapentate, indometacin farnesil, teprenone, and tocopherol nicotinate). The total daily drug dose administered in these formulations, which range in complexity from simple solutions of the drug in a dietary oil up to multi-excipient, self-emulsifying drug delivery systems (SEDDS), range from less than 0.25 μg to greater than 2000 mg. The amount of drug contained in a unit-dose capsule product ranges from 0.25 μg to 500 mg and for oral solution products, from 1 μg/mL to 100 mg/mL. The total amount of lipid excipient administered in a single dose of a capsule formulation ranges from 0.5 to 5 grams, but can range from as low as 0.1 mL to as high as 20 mL for oral solution products. Some of these products tolerate room temperature storage for only brief periods of time and require long-term storage at 2–8 °C due to chemical and/or physical stability issues.
Table 1.5: List of Selected Commercially Available Lipid-based Formulations for Oral Administration in the United States in 2005

<table>
<thead>
<tr>
<th>Molecule/trade name/company</th>
<th>Indication</th>
<th>Dose</th>
<th>Type of Formulation/Strength</th>
<th>Lipid excipients and surfactants</th>
<th>Nonlipid excipients</th>
<th>Storage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Amprenavir/ Agenerase®/ GlaxoSmithKline</td>
<td>HIV antiviral</td>
<td>1200 mg (8 capsules) b.i.d</td>
<td>Soft gelatin capsule, 50, 150 mg</td>
<td>TPGS (280 mg in the 150 mg capsule)</td>
<td>PEG 400 (247, 740 mg), propylene glycol (19.57 mg)</td>
<td>RT</td>
</tr>
<tr>
<td>Baxarotene/ Targetin®/ Ligand</td>
<td>Antineoplastic</td>
<td>300-750 mg (4-10 capsules) q.d.</td>
<td>Soft gelatin capsule, 75 mg</td>
<td>Polysorbate 20</td>
<td>PEG 400, povidone, BHA</td>
<td>RT, avoid high temp, humidity and light</td>
</tr>
<tr>
<td>Calcitriol/ Rocaltrol®/ Roche</td>
<td>Calcium regulator</td>
<td>Adults: 0.25-0.5 mcg (1 capsule) q.d.</td>
<td>Soft gelatin capsule, 0.25-0.5 mcg</td>
<td>Fractionated triglyceride of coconut oil (MCT)</td>
<td>BHA, BHT</td>
<td>15-30°C protect from light</td>
</tr>
<tr>
<td>Ciprofloxacin/ Cipro®/ Bayer</td>
<td>Antibiotic</td>
<td>15 mg/kg b.i.d. not to exceed the adult dose of 500 mg per dose</td>
<td>Microcapsules for constitution to suspension, 5 % or 10 % in solid, 50 or 100 mg/ml in suspension</td>
<td>Bottle 1- diluents: MCT, sucrose, lecithin, water and strawberry flavour</td>
<td>Bottle 1- Solid: PVP, methacrylic acid copolymer, HPMC, magnesium stearate and Polysorbate 20</td>
<td>Store below 30°C but not frozen</td>
</tr>
<tr>
<td>Cyclosporin A/I. Neoral®/Novartis</td>
<td>Immunosuppressant/Prophylaxis for organ transplant rejection</td>
<td>2-10 mg/kg/day, b.i.d. (1-7 capsules)</td>
<td>Soft gelatin capsule, 10, 25, 50, 100 mg</td>
<td>dl-α-tocopherol, corn oil-mono-di-triglycerides, cremophore RH 40</td>
<td>Ethanol 11.9%, glycerol, propylene glycol</td>
<td>RT</td>
</tr>
<tr>
<td>Drug</td>
<td>Dosage</td>
<td>Formulation</td>
<td>Excipients</td>
<td>Storage</td>
<td></td>
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</tr>
<tr>
<td>Cyclosporin A/II. Sandimmune®/Novatis</td>
<td>2-10 mg/kg/day, b.i.d. (1-7 ml)</td>
<td>Oral Solution 100 mg/ml</td>
<td>dl-α-tocopherol, corn oil-monodi-triglycerides, cremophore RH 40</td>
<td>Ethanol 11.9%, glycerol, propylene glycol</td>
<td>RT, do not store in the refrigerator</td>
<td></td>
</tr>
<tr>
<td>Cyclosporin A/III Gengraf®/Abbott</td>
<td>2-10 mg/kg/day, b.i.d. (1-7 ml)</td>
<td>Soft gelatin capsule, 25, 100 mg</td>
<td>Corn oil, Labrafil M-2125CS</td>
<td>Ethanol 12.7%, glycerol</td>
<td>RT</td>
<td></td>
</tr>
<tr>
<td>Cyclosporin A/IV Cyclosporin®/Sidmak</td>
<td>2-10 mg/kg/day, b.i.d. (1-7 capsules)</td>
<td>Hard gelatin capsule, 25, 100 mg</td>
<td>Cremophor EL, Polysorbate 80</td>
<td>Ethanol 12.8%, propylene glycol</td>
<td>RT</td>
<td></td>
</tr>
<tr>
<td>Droxercalciferol/ Hectorol® Bone care</td>
<td>1-9 mg/kg/day 70-700 mg, (1-7 capsules)</td>
<td>Soft gelatin capsule, 100 mg</td>
<td>Labrafac, dl-α-tocopherol glyceryl caprylate, Labrasol, Cremophor EL.</td>
<td></td>
<td>RT</td>
<td></td>
</tr>
<tr>
<td>Dronabinol/Marino®/ Roxane and Unimed</td>
<td>2.5-10 mg (1 capsule) b.i.d.</td>
<td>Soft gelatin capsule 2.5, 5, 10 mg</td>
<td>Sesame oil</td>
<td>None</td>
<td>8-15°C protect from freezing</td>
<td></td>
</tr>
<tr>
<td>Dutasteride/Avodart® / GlaxoSmithKline</td>
<td>0.5 mg q.d. (1 capsule)</td>
<td>Soft gelatin capsule, 0.5 mg</td>
<td>Mixture of mono- and diglycerides of caprylic/capric acid</td>
<td>BHT</td>
<td>RT</td>
<td></td>
</tr>
<tr>
<td>Isotretinoin/Accutane®/ Roche</td>
<td>0.5-1.0 mg/kg/day subdivided in two doses (1-2 capsules)</td>
<td>Soft gelatin capsule, 10, 20, 40 mg</td>
<td>Beeswax, hydrogenated soybean oil flakes, hydrogenated vegetable oils, soybean oil</td>
<td>BHA, EDTA</td>
<td>RT, protect from light</td>
<td></td>
</tr>
<tr>
<td>Lopinavir and ritonavir/Kaletra® Abbott</td>
<td>400/100 mg b.i.d. (2 tablets) or 800/200 mg q.d. (4 tablets)</td>
<td>Tablet 200 mg lopinavir and 50 mg ritonavir</td>
<td>Sorbitan monolaurate</td>
<td>Sodium stearyl fumarate, crosspovidone and silicon dioxide</td>
<td>RT</td>
<td></td>
</tr>
<tr>
<td><strong>Chapter 1</strong></td>
<td><strong>Introduction</strong></td>
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</tr>
<tr>
<td>400/100 mg b.i.d. (3 capsules)</td>
<td>Soft gelatin capsule, 133.3 mg lopinavir and 33.3 mg ritonavir</td>
<td>Oleic acid, Cremophor EL</td>
<td>Propylene glycol</td>
<td>2-8°C, or at RT for ≤ 2 months</td>
<td></td>
<td></td>
</tr>
<tr>
<td>400/100 mg b.i.d. (5 ml)</td>
<td>Oral Solution, 80 mg/ml lopinavir and 20 mg/ml ritonavir</td>
<td>Cremophor RH 40, peppermint oil</td>
<td>Alcohol (42.2% v/v), glycerine, propylene glycol, sodium chloride, sodium citrate, citric acid, water, flavours/sweeteners</td>
<td>2-8°C, or at RT for ≤ 2 months</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Progesterone/Prometrium®/Solvay</td>
<td>Hormone replacement therapy</td>
<td>Soft gelatin capsule, 100, 200 mg micronized</td>
<td>Peanut oil</td>
<td>None</td>
<td>RT, protect from light and excessive moisture</td>
<td></td>
</tr>
<tr>
<td>Ritonavir/Norvir®/Abbott</td>
<td>HIV antiviral</td>
<td>Adults 600 mg (6 capsules) b.i.d.</td>
<td>Soft gelatin capsule, 100 mg</td>
<td>Oleic acid, Cremophor EL</td>
<td>BHT, Ethanol</td>
<td>2-8°C, or at RT for ≤ 1 months</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Pediatrics 250-450 mg/m² up to a max. Of 600 mg (&lt;7.5 ml) b.i.d.</td>
<td>Oral Solution, 80 mg/ml</td>
<td>Cremophore EL, sweetener, dye</td>
<td>Ethanol (43%), water, propylene glycol, citric acid, flavours</td>
<td>RT</td>
</tr>
<tr>
<td>Saquinavir/Fortovase®/Roche</td>
<td>HIV antiviral</td>
<td>1200 mg capsule (6 capsules) t.i.d. without ritonavir, 1000 mg (5 capsules) b.i.d. with ritonavir</td>
<td>Soft gelatin capsule, 200 mg</td>
<td>Medium chain mono- and diglycerides, dl-a-tocopherol</td>
<td>Povidone</td>
<td>2-8°C, or at RT for ≤ 3 months</td>
</tr>
<tr>
<td>Sirolimus/Rapamune®/Wyeth-Ayerst</td>
<td>Immuno-suppressant</td>
<td>6 mg (6 ml) loading dose followed by 2 mg (2ml) q.d.</td>
<td>Oral Solution, 1 mg/ml</td>
<td>Phosal 50 PG (phosphatidyl choline, mono-and diglycerides, soy fatty acids, ascorbyl palmitate), polysorbate 80</td>
<td>Phosal 50 PG, propylene glycol, ethanol (1.5-2.5%)</td>
<td>2-8°C, or at RT for &lt; 15 days</td>
</tr>
<tr>
<td>Tipranavir/Aptivus®/Boehringer Ingelheim</td>
<td>HIV antiviral</td>
<td>500 mg (2 capsules) with ritonavir 200 mg b.i.d.</td>
<td>Soft gelatin capsule, 200 mg</td>
<td>Cremophor EL, medium chain mono- and diglycerides</td>
<td>Ethanol (7% w/w or 0.1 g per capsule), propylene glycol</td>
<td>2-8°C prior to opening the bottle, RT for &lt; 60 days</td>
</tr>
<tr>
<td>Tolterodine tartarate/Detrol LA® Pharmacia &amp; UpJohn</td>
<td>Overactive bladder-muscarinic receptor antagonist</td>
<td>2-4 mg q.d. (1 capsule)</td>
<td>Extended release hard gelatin capsule, 2, 4 mg</td>
<td>MCT, oleic acid</td>
<td>Sucrose, hypromellose, ethylcellulose</td>
<td>RT</td>
</tr>
<tr>
<td>Tretinoin/Vesanoid®/Roche</td>
<td>Antineoplastic</td>
<td>45 mg/m² subdivided (8 capsules) b.i.d.</td>
<td>Soft gelatin capsule, 10 mg</td>
<td>Beeswax, hydrogenated soybean oil flakes, hydrogenated vegetable oils, soybean oil</td>
<td>BHA, EDTA</td>
<td>RT</td>
</tr>
<tr>
<td>Molecule/trade name/company</td>
<td>Indication</td>
<td>Dose</td>
<td>Type of Formulation/Strength</td>
<td>Lipid excipients and surfactants</td>
<td>Nonlipid excipients</td>
<td>Storage</td>
</tr>
<tr>
<td>----------------------------</td>
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</tr>
<tr>
<td>Alfacalcidol/One-Alpha® capsule/Leo Laboratories</td>
<td>Calcium regulator</td>
<td>0.5-1 mcg q.d. (1 capsule)</td>
<td>Soft gelatin capsule, 0.25, 0.5, 1.0 µg</td>
<td>Sesame oil, dl-α-tocopherol</td>
<td>None</td>
<td>RT</td>
</tr>
<tr>
<td>Clofazimine/Lamprene® Capsule 100 mg/ Alliance Pharmaceuticals</td>
<td>Treatment of leprosy in combination with dapsone and rifampicin</td>
<td>Maximum 300 mg q.d. for upto 3 months (3 capsules)</td>
<td>Soft gelatin capsule, 100 mg (micronized suspension in an oil-wax base)</td>
<td>Rapeseed oil, wax blend (beeswax hydrogenated soybean oil, partially hydrogenated plant oils)</td>
<td>BHT, citric acid, PG</td>
<td>Below 25°C</td>
</tr>
<tr>
<td>Clomethiazole edisilate/Heminevrin® Capsules/AstraZeneca</td>
<td>Sedative</td>
<td>1-4 capsules as needed</td>
<td>Soft gelatin capsule, 192 mg</td>
<td>MCT from fractionated coconut oil</td>
<td>None</td>
<td>RT</td>
</tr>
<tr>
<td>Efavirenz/Sustiva® Oral Solution/Bristol-Meyers Squibb</td>
<td>HIV antiviral</td>
<td>Adults: 600 mg q.d. (up to 20 ml) Pediatrics: 270-600 mg (9-20 ml)</td>
<td>Oral Solution, 30 mg/ml</td>
<td>MCT</td>
<td>Benzoic acid, strawberry/mint flavour</td>
<td>RT</td>
</tr>
<tr>
<td>Fenofibrate/Fenogal®/Genus</td>
<td>Anti-hyperlipoproteine mic</td>
<td>200 mg (1 capsule) q.d.</td>
<td>Hard gelatin capsule, 200 mg</td>
<td>Gelucire 44/14, hydrogenated vegetable oil</td>
<td>PEG-20,000, HPC</td>
<td>RT</td>
</tr>
<tr>
<td>Morphine sulphate/ MXL® capsules/Napp Pharmaceuticals</td>
<td>Analgesic</td>
<td>30-200 mg q.d. (1 capsule)</td>
<td>Hard gelatin capsule containing multiparticulates, 30, 60, 90, 120, 150 and 300 mg</td>
<td>Hydrogenated vegetable oil</td>
<td>PEG-6000, talc, magnesium stearate</td>
<td>RT</td>
</tr>
<tr>
<td>Molecule/trade name/company</td>
<td>Indication</td>
<td>Dose</td>
<td>Type of Formulation/Strength</td>
<td>Lipid excipients and surfactants</td>
<td>Nonlipid excipients</td>
<td>Storage</td>
</tr>
<tr>
<td>----------------------------</td>
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</tr>
<tr>
<td>Testosterone undecanoate/Restandol® 40 mg/Organon Laboratories</td>
<td>Hormone replacement therapy</td>
<td>40-160 mg q.d. (1-4 capsules) undecanoate</td>
<td>Soft gelatin capsules, 40 mg HSE (equivalents of testosterone, 61 mg of testosterone)</td>
<td>Oleic acid</td>
<td>None</td>
<td>2-8°C until dispensed, then at RT after dispensing.Protect from light and heat</td>
</tr>
<tr>
<td>Valproic acid/Convulex® 100 mg, 200 mg, 500 mg/Pharmacia</td>
<td>Antiepileptic</td>
<td>10-60 mg/kg/day upto 2500 mg per day (1-5 capsules)</td>
<td>Soft gelatin capsule, 100, 200 and 500 mg</td>
<td>MCT</td>
<td>HPMC-phthalate and dibutylphthalate</td>
<td>RT</td>
</tr>
</tbody>
</table>

**Table 1.7: List of Selected Commercially Available Lipid-based Formulations for Oral Administration in the Japan in 2005**
<table>
<thead>
<tr>
<th>Capsules 15 mg/Eisai Co.</th>
<th>capsules) t.i.d.</th>
<th>capsule are a viscous liquid or semisolid</th>
<th>esters of fatty acid, glyceryl monooleate</th>
<th>protect from light</th>
</tr>
</thead>
<tbody>
<tr>
<td>Teprenone/Selbex® capsules 50 mg and fine granules 10%/Eisai Co.</td>
<td>Acute gastritis 150 mg (3 capsules) t.i.d.</td>
<td>Hard capsule, 50 mg (contents of capsule are granules or powder)</td>
<td>α-tocopherol</td>
<td>Hydrated silicon dioxide, talc, mannitol, PEG 6000, lactose</td>
</tr>
<tr>
<td></td>
<td>150 mg (1.5 g granules) t.i.d.</td>
<td>Fine granules, 10% w/w</td>
<td>α-tocopherol</td>
<td>Hydrated silicon dioxide, talc, HPMC, mannitol, lactose</td>
</tr>
<tr>
<td>Tocopherol nicotinate/ Juvela® N Soft Capsules/Eisai Co.</td>
<td>Hypertension, hyperlipidemia 200 mg (1 capsule) t.i.d.</td>
<td>Soft gelatin capsule, 200 mg (contents of capsule are a viscous suspension or semisolid)</td>
<td>MCT, glycol esters of fatty acid</td>
<td>Aspartic acid</td>
</tr>
</tbody>
</table>
1.10. OUTLINE OF THE STUDY

The present research work is focused on exploiting a few surfactants/and or solubilizers to develop self-emulsifying drug delivery system for two investigational lipophilic model drugs with various ratios of different excipients, commonly used in lipid-based drug delivery system. The work also includes the pharmacokinetic study of all the developed formulations in animal model.

The research work is divided into two Sections, viz. **Section I** and **Section II**, dealing with the two investigational model drugs **Nevirapine** and **Itraconazole**, respectively. For convenience, the formulation development for both the drugs is described into two parts.

First part of both the sections (**Chapter 2 and 4**) deals with the SEDDS development using Soluphor® P. Chemically, it is a 2-pyrrolidine (2P) and has been widely used for topical (Femenia-Font et al., 2006) and parenteral (Shah and Agnihotri, 2011) pharmaceutical dosage forms. Few literatures are also available on human oral use of 2-pyrrolidone for improvement of solubility and dissolution of poorly water soluble drugs (Khachane et al., 2011; Schamp et al., 2006). Physicochemical properties, endogenous origin and toxicity of Soluphor® P are mentioned in introduction of **Chapter 2**. Several drug delivery systems mostly focused on the delivery of oral dosage forms and injectables (biodegradable implants) using Soluphor® P has been reviewed in the introductory part of the **Chapter 4**. The introduction section of **Chapter 4** also discussed N-methyl-2-pyrrolidone, which is widely used as a solubilizer in the development of various pharmaceutical dosage forms and compared its physicochemical properties and toxicity issues with that of Soluphor® P (2-pyrrolidone). The present research work has explored the ability of Soluphor® P to improve the solubility; permeability and bioavailability of BCS class II drugs.

Second part of both the sections (**Chapter 3 and 5**) reveal the classical approach for the development of self emulsifying drug delivery system of poorly water soluble drugs using traditional excipients. For this, surfactants like Tween 20, Tween 80, Labrasol® and co-surfactant Transcutol P have been used.

**Chapter 3** describes that, patients infected with HIV experience a variety of functional and anatomical abnormalities in the gastrointestinal tract that result in diarrhoea and
nutrient malabsorption of fat, carbohydrates, specific micronutrients and proteins. In such situations, medium chain triglycerides (MCT) are readily absorbed from the small bowel under conditions in which the absorption of long chain triglycerides (LCT) is impaired (Griffin, 1990; Wanke et al., 1996). It has been also investigated that dietary formulations providing majority of fat in the form of MCT have resulted in the improvement of diarrhoea, intestinal discomfort, bloating and nitrogen balance both in children and adults with chronic diarrhoea from chronic pancreatitis, short bowel syndrome, non-specific enteropathy, cystic fibrosis and biliary atresia (Bach & Babayan, 1982; Wanke et al., 1996).

Chapter 5 addresses the hydrophobicity of a broad spectrum antifungal agent, Itraconazole and the need of acidic conditions of GIT for its better absorption. Co-administration of acidic beverage has been shown to improve the absorption of Itraconazole especially those who have hypochlorhydria or achlorhydria.
REFERENCES


Chapter 1

Introduction


SECTION I
Nevirapine
Page no. 43 to 55
I.1. Drug Profile

**NEVIRAPINE**

(Indian Pharmacopoeia, Addendum 2005; Merck Index, 2006; Moffat et al., 2004; Physician Desk Reference, 2008; Raffanti and Haas, 2001; Sweetman, 2005)

![Figure I.1. Structure of Nevirapine](image)

**Chemical Formula:** C\(_{15}\)H\(_{14}\)N\(_4\)O  
**Molecular weight:** 266.30  
**CAS name:** 11-Cyclopropyl-5,11-dihydro-4-methyl-6H-dipyrido(3,2-b:2,3-e)(1,4)diazepin-6-one  
**CAS registry No.:** 129618-40-2

**Physicochemical properties:**

**Description:** White to off-white crystalline powder  
**Solubility:** Soluble in dimethyl sulfoxide (DMSO), sparingly soluble in dichloromethane (DCM) and in dimethylformamide (DMF); highly soluble in water at pH < 3 but solubility decreases to approximately 0.1 g/L at neutral pH.  
**Dissociation constant (pKa):** 2.8  
**Polarity/partition coefficient (log P):** 2.5  
**Melting point:** 242 - 246°C

**Mechanism of action:**

Nevirapine (NVP) is a non-nucleotide reverse transcriptase inhibitor (NNRTI). It binds directly to reverse transcriptase (RT) and blocks the RNA dependent and DNA dependent DNA polymerase activities by causing a disruption of the enzyme’s catalytic site. The activity of nevirapine does not compete with template or nucleoside triphosphates. HIV-2 RT and eukaryotic DNA polymerases (such as human DNA polymerases α, β, γ, δ) are not inhibited by nevirapine.
Antiviral activity:

The antiviral activity of nevirapine has been measured in a variety of cell lines including peripheral blood mononuclear cells, monocyte derived macrophages, and lymphoblastoid cell lines. EC$_{50}$ values (50% inhibitory concentration) ranged from 14-302 nM against laboratory and clinical isolates of HIV-1, using human cord blood lymphocytes and human embryonic kidney 293 cells. Nevirapine in combination with efavirenz exhibited strong antagonistic anti-HIV-1 activity in cell culture and was additive to the antagonistic with protease inhibitor ritonavir or the fusion inhibitor enfuvirtide. Nevirapine exhibited additive to synergistic anti-HIV-1 activity in combination with the protease inhibitors amprenavir, atazanavir, indinavir, lopinavir, nelfinavir, saquinavir and tipranavir, and the NRTIs abacavir, didanosine, emtricitabine, lamivudine, stavudine, tenofovir and zidovudine. The anti-HIV-1 activity of nevirapine was antagonized by anti-HBV drug adefovir, and by the anti-HCV drug ribavirin in cell culture.

HIV-1 isolates with reduced susceptibility (100-250-fold) to nevirapine emerge in cell culture. Genotypic analysis showed mutations in the HIV-1 RT gene Y181C and/or V106A depending upon the virus strain and cell line employed. Time to emergence of resistance in cell culture was not altered when selection included nevirapine in combination with several other NNRTIs.

Pharmacokinetics

Absorption: Nevirapine is readily absorbed (> 90%) after oral administration in healthy volunteers and in adults with HIV-1 infection. Absolute bioavailability in 12 healthy adults following single-dose administration was 93 ± 9% for a 50 mg tablet and 91 ± 8% for an oral solution. Peak plasma Nevirapine concentrations of 2 ± 0.4 µg/mL were attained by 4 hours following a single 200 mg dose.

Distribution: Nevirapine is highly lipophilic and essentially non-ionized at physiological pH. Following intravenous administration to healthy adults, the apparent volume of distribution (V$_d$) of nevirapine was 1.21 ± 0.09 L/kg, suggesting that nevirapine is widely distributed in humans. It readily crosses the placenta and is also found in breast milk. Nevirapine is about 60% bound to plasma proteins in the plasma concentration range of 1-10 µg/mL. Nevirapine concentration in human cerebrospinal fluid (n=6) were 45% (± 5%) of the concentrations in plasma. This ratio is approximately equal to the fraction not bound to plasma protein.
Metabolism/Elimination: *In vivo* studies in human and *in vitro* studies with human liver microsomes have shown that nevirapine is extensively biotransformed via cytochrome P450 (oxidation) metabolism to several hydroxylated metabolites. *In vitro* studies with human liver microsomes suggest that oxidative metabolism of nevirapine is mediated primarily by cytochrome P450 (CYP) isozymes from the CYP3A4 and CYP2B6 families, although other isozymes may have a secondary role. In a mass/balance excretion study in 8 healthy male volunteers dosed to steady state with nevirapine 200 mg given twice daily followed by single 50 mg dose of \(^{14}\)C-nevirapine, approximately 91.4 ± 10.5 % of the radio-labelled dose was recovered, with urine (81.3 ± 11.1 %) representing the primary route of excretion compared to feces (10.1 ± 1.5%). Greater than 80% of the radioactivity in urine was made up of glucouronide conjugates of hydroxylated metabolites. Thus cytochrome P450 metabolism, glucouronide conjugation and urinary excretion of glucourinated metabolites represent the primary route of nevirapine biotransformation and elimination in humans. Nevirapine is an inducer of hepatic cytochrome P450 metabolic enzymes 3A4 and 2B6. It induces CYP3A4 and CYP2B6 by approximately 20-25%, as indicated by erythromycin breath test results and urine metabolites. Autoinduction of CYP3A4 and CYP2B6 mediated metabolism leads to an approximately 1.5 to 2 fold increase in the apparent oral clearance of nevirapine as treatment continues form a single dose to two-to-four weeks dosing with 200-400 mg/day. Autoinduction also results in a corresponding decrease in the terminal phase half-life of nevirapine in plasma, from approximately 45 hours (single dose) to approximately 25-30 hours following multiple dosing with 200-400 mg/day.

Interaction: Nevirapine being itself mild to moderate enzyme inducer may thus reduce plasma concentrations of other drugs.

Antibacterials: Plasma concentrations of nevirapine may be decreased by rifabutin and rifampicin, probably as a result of enzyme induction.

Antifungal: Use of nevirapine and ketoconazole may result in reduction in the plasma concentration of ketoconazole and increase in that of nevirapine. Use of nevirapine with fluconazole may increase the bioavailability of nevirapine and caution required when the drugs are used together.

Hormonal contraceptives: Nevirapine may decrease the plasma concentrations of hormonal contraceptives.
Adverse effects: The most frequently adverse events are rash, nausea, fatigue, somnolence, headache and abnormal liver function test. Rashes are usually mild to moderate, maculopapular erythematous cutaneous eruptions, with or without pruritus, located on the trunk, face and extremities. Allergic reactions (anaphylaxis, angioedema and urticaria) have been reported. Severe and life-threatening skin reactions have occurred in patients treated with nevirapine, including Steven-Johnson syndrome (SJS) and toxic epidermal necrolysis (TEN). Rashes occur alone or in the context of hypersensitivity reactions characterized by rash with constitutional symptoms such as fever, arthralgia, myalgia and lymphadenopathy plus visceral involvement, such as hepatitis, eosinophilia, granulocytopenia and renal dysfunction.

I.2. Background of the study
Introduction

Nevirapine, chemically 11-cyclopropyl-5,11- dihydro-4-methyl-6H-dipyrido[3,2-b: 2’,3’-e] diazepin- 6-one, a dipyridodiazepinone (Murphy and Montaner, 1996; Malaty and Kupper, 1999) is a non-nucleoside reverse transcriptase inhibitor of human immunodeficiency virus type 1 (HIV-1). Nevirapine was discovered by Hargrave et al. (1994) at Boehringer Ingelheim Pharmaceuticals, Inc. It is covered by US patent and corresponding foreign patents. Nevirapine was the first Non-Nucleoside Reverse Transcriptase Inhibitor (NNRTI) approved by the FDA of the United States and currently used in the treatment of HIV-1 infections (Waters et al., 2007; Devi and Pai, 2006). It was approved on June 21, 1996 for adults and September 11, 1998 for children. It was also approved in Europe and Canada in 1997 and 1998, respectively.

It is a hydrophobic drug with a log octanol–water partition coefficient (log \( P \)) = 2.5, \( T_m = 244.5^\circ C \), and pKa = 2.8 (Wishart et al., 2006; Pereira et al., 2007) and is practically insoluble in water (0.1 mg/mL) at physiological pH conditions and soluble only under extremely acidic media (Merck Index, 2006). Anhydrous form of nevirapine is used in tablet formulations, while nevirapine hemihydrate is used in suspension formulation (AHFS Drug Info, 2004). Although, the drug appears to be readily absorbed orally, nevirapine particularly at higher doses (> 50 mg) exhibit characteristics of solubility rate-limited absorption with a resultant decrease in bioavailability (Hawi and Bell, 1994; Lamson et al., 1995). Nevirapine is also used in fixed dose combinations (FDCs) along with lamivudine and stavudine. In comparison to other drugs, nevirapine (NVP) is poorly
soluble hydrophobic molecule which poses processing problems in the manufacture of FDCs (Sarkar et al., 2008).

Nevirapine belongs to Biopharmaceutical Classification System (BCS) class II (low solubility/high permeability), poses a challenge in achievement of optimal dissolution kinetics from the dosage form (Kasim et al., 2004). NVP is a weak base (pKa= 2.8) with low intrinsic water solubility (0.06 mg/ml) which gives rise to difficulties in the formulation of dosage forms and leads to variable dissolution rates with a resultant decrease in bioavailability (Hawi and Bell, 1994; Macha et al., 2009a and 2009b; Lamson et al., 1995).

**HIV Infection:**

At the present time, more than 33 million people are infected with the human immunodeficiency virus (HIV), with 2.5 million new infections diagnosed in 2008. Sub-Saharan Africa remains most heavily affected by the pandemic, accounting for 67% of all people living with HIV and >70% of deaths from the acquired immune deficiency syndrome (AIDS) in recent years. Globally, the percentage of women among people living with HIV has remained stable at 50% for several years, but women’s share of infection is increasing in several countries. The overall number of people living with HIV has increased as a result of new infections and the beneficial effects of more widely available antiretroviral therapy (HIV/AIDS, 2008). The introduction of combination antiretroviral therapy, compounded with the routine use of HIV RNA viral load and CD4+ T-cell counts as surrogate markers of drug efficacy and disease progression (Mellors et al., 1996), has brought about a dramatic increase in life expectancy among HIV-infected patients (The-Antiretroviral-Therapy-Cohort-Collaboration, 2008).

There have been significant accomplishments in the past 25 years in terms of greater emphasis on disease prevention, technologies for diagnosis, and development of innovative therapeutic strategies (Gallo, 2006). At present, there are over 20 different antiretroviral drugs approved in the United States under the general classes of nucleoside reverse transcriptase inhibitors (NRTI), non-nucleoside reverse transcriptase inhibitors (NNRTI), protease inhibitors (PI), and fusion inhibitors (FI) (Chearskul et al., 2006).

Current treatment options have not yet reached the vast majority of people living with HIV. Although the eradication of HIV infection is unlikely to be a short-term prospect, the voices of experts have begun to herald a change in the future paradigm of HIV treatment
by shifting the new long-term goal towards achieving virus control that would allow for durable drug-free remissions (Richman et al., 2009). HIV infection cannot be cured with current treatments and patients are destined to undergo treatment for life. Simplification of therapy, improvement of patient adherence and minimization of drug resistance are foreseeable near-term goals that will likely also be important stepping stones towards achieving long-term drug-free virus control and ultimately finding a cure.

One of the major problems in the chronic treatment is the fact that the viral particles are able to reside in cellular and anatomical sites in the body following replication and remain viable even when there are adequate drug concentrations in the blood (Schrager and D’Souza, 1998; Chun et al., 2000). Examples of cellular reservoirs include T-lymphocytes, monocytes, and macrophages, while the major anatomical reservoirs include central nervous system (CNS), lymph nodes, liver, spleen, lungs, and the genitals (Vyas et al., 2006). Poor drug availability in the cellular and anatomical reservoirs is affected by expression of efflux transporters (e.g., P-glycoprotein), presence of drug metabolizing enzymes (e.g., cytochrome P-450), poor permeability properties, non-targeted distribution, and rapid clearance. The reduced bioavailability and short residence of anti-retroviral agents at these viral reservoir sites have profound impact on the clinical management of the disease. The overall consequence is that upon discontinuation of therapy or when drug resistance develops, HIV is able to re-seed the systemic circulation and continue to propagate the infection (Clarke et al., 2000, Kulkovsky and Bray, 2006).

The immunopathogenesis of HIV/AIDS has been previously amply documented; from the time of infection to the end stage of the disease (Chinen and Shearer, 2008). The end stage of the disease may be characterised by a spectrum of diseases (Stoddart and Reyes, 2006) including opportunistic infections (such as Pnuemocystis carinii and Mycobacteruim tuberculosis), dementia and cancer (Stoddart and Reyes, 2006). In addition to macrophages, lymph nodes, bone marrow, spleen and lungs, the CNS represents one of the most important anatomical sites of the virus after infection. This causes significant neuronal damage and loss that often leads to HIV associated dementia. Without treatment, HIV 1 infection is nearly uniformly fatal within 5-10 years (Stoddart and Reyes, 2006).

The majority of orally administered drugs gain access to the systemic circulation by absorption into the portal blood. However, for some extremely lipophilic compounds, transport via the intestinal lymphatics provides an additional route of access to the systemic circulation. Exogenous compounds absorbed via the intestinal lymph are
generally transported in association with the lipid core of intestinal lipoprotein thereby requiring co-administered lipid to stimulate lipoprotein formation.

B- and T- lymphocytes, which play a major role in maintaining the immune system, also circulate through the lymphatics in relatively high concentrations compared with systemic blood. Consequently, there is considerable interest in the specific delivery of anti-infective or anti-viral agents to the lymph to combat lymphocyte destruction by, for example, HIV (Porter and Charman, 1997).

I.3. Scope and objectives of the present work

Reducing the dosing frequency would significantly improve patient compliance and their quality of life. To study new forms of administration, it is necessary to do pre-clinical studies and know the absorption characteristics of nevirapine in laboratory animals. However, there are no reports about its bioavailability in rats and about its pharmacokinetic. One of the objectives of this study was to describe the pharmacokinetics of nevirapine in rats after oral administration of the developed SEDDS and marketed suspension.

Although the metabolism of NVP in different animal species has been studied (Riska et al., 1999), pharmacokinetic studies in experimental animals are few, and there is meagre information available about the bioavailability of this drug in laboratory animals, e.g. in rats (Usach and Peris, 2011). Knowledge on the absorption characteristics of NVP in laboratory animals is crucial for undertaking pre-clinical studies on new dosage forms to reduce the dose or dosing frequency. Since HIV-infected patients require long-term therapy with antiretroviral drugs, such as NVP, an improvement in the treatment regimen will improve their quality of life. This objective could be reached through various dosage delivery approaches and the self-emulsifying drug delivery system is one of them. A rash which is the major adverse reaction and few others can be minimized if the dose can be reduced.

A range of novel strategies are currently being developed for efficient delivery of anti-retroviral (ARV) drugs including nevirapine. Efficient delivery could be achieved by encapsulating the drug or by attaching it with a carrier system (Ferrari, 2005; Duncan, 2003; Gonzalez de Requena et al., 2002). Several delivery systems have been reported for the delivery of ARV drugs including bioadhesive coated matrix tablets (Betagiri et al., 2001; Govender et al., 2005), ceramic implants (Benguzzi 2000), liposomes
Section I

Nevirapine

(Desormeaux and Bergeron, 1998; Jain et al., 2006; Jin et al., 2005; Makabi Panzu et al., 1998; Ramana et al., 2010), solid colloidal nanoparticles (Mainardes et al., 2009; Kuo and Su, 2007; Chattopadhyay et al., 2008; Kaur et al., 2008), microparticles by supercritical antisolvent method (Sanganwar et al., 2010), dendrimers (Dutta et al., 2007), micelles & microemulsion (Griffin and Driscoll, 2006), nanopowders (Erdenburgh et al., 2007) and suspensions/nanosuspension (Kinman et al., 2003; Shegokar and Singh, 2011; Shegokar et al., 2011; Yang et al., 2011). Table I.1 shows the commercially available nevirapine formulations in India.

Table I.1: Commercially available nevirapine formulations in India

<table>
<thead>
<tr>
<th>Brand Name</th>
<th>Dosage form</th>
<th>Company</th>
</tr>
</thead>
<tbody>
<tr>
<td>Neve</td>
<td>Tablet</td>
<td>Cadila</td>
</tr>
<tr>
<td>Nevimune</td>
<td>Tablet, Suspension</td>
<td>Cipla</td>
</tr>
<tr>
<td>Nevipan</td>
<td>Tablet</td>
<td>Ranbaxy</td>
</tr>
<tr>
<td>Neviretro 200</td>
<td>Tablet</td>
<td>Alkem</td>
</tr>
<tr>
<td>Nevivir</td>
<td>Tablet</td>
<td>Hetro HC</td>
</tr>
</tbody>
</table>

The main objective of the study was to contribute to the understanding of the physicochemical principles, key factors in predicting the performance and applicability of self-emulsifying drug delivery system for the improvement of dissolution and in vivo performance of poorly water soluble/lipophilic drugs.

The specific objectives of this investigation include:

1. To explore SEDDS as a novel formulation for a lipophilic drug- nevirapine
2. This novel SEDDS formulation for nevirapine will be suitably modified so as to
   a. Enhance the solubility
   b. Increase diffusion and permeation of the drug
3. To compare the new formulations thus developed with marketed formulation for their in vivo absorption using a suitable animal model
4. In addition, to see the effect of ratios of oil and surfactant on droplet size
5. To study the effect of droplet size on uptake of the drug by gastrointestinal tissue of rat
REFERENCES


Section I

Nevirapine

performance of sucrose co-freeze dried solid nanoparticulate powder of the anti-HIV agent


Optimisation and characterization of bioadhesive controlled release tetracycline

Griffin, B.T., Driscoll, C.M.O., 2006. A comparison of intestinal lymphatic transport and
systemic bioavailability of saquinavir from three lipid based formulation in the

Hargrave, K.D., Proudfoot, J.R., Adams, J., Grozinger, K.G., Schmidt, G., Engle, W.,
Trummlitz, G., Eberlein, W., 1994. 5,11-dihydro-6H-dipyrido (3,2-B: 2’,3’-E)(1,4)
diazepines and their use in the prevention or treatment of HIV infection. U.S. Patent
5366972.

Hawi, A., Bell, G., 1994. Preformulation studies of nevirapine, a reverse transcriptase
inhibitor. *Pharm. Res.* 11 (Suppl), S 236


tissue distribution of zidovudine in rats following intravenous administration of

Kasim, N.A., Whitehouse, M., Ramachandran, C., Bermejo, M., Lennernas, H., Hussain,


Section I

Nevirapine


