Sildenafil citrate nanoemulsion vs. self-nanoemulsifying delivery systems: rational development and transdermal permeation

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Abstract: Sildenafil citrate (SC) is the first choice drug for erectile dysfunction. Nevertheless, drug oral delivery is hampered by some obstacles including first pass metabolism, numerous side effects, relatively short duration and long onset of action. Furthermore, drug delivery formulation and transdermal application of SC is challenged by its amphoteric nature, low oil and water solubility, pH-dependent characteristics and poor membrane permeability. In this paper, relevance of nanomedicine to improve SC characteristics and transdermal permeation was assessed. SC-loaded self-nanoemulsifying drug delivery system (SNEDDS) and nanoemulsions have been developed and appraised. Both nanocarriers encompassed the bioactive excipient, Cremophor RH40® as a surfactant. The nanocarriers encompassed an oil blend of Caproyl 90® and Maisine 35-1® and propylene glycol as a co-surfactant. Nanocarrier assessment was based on solubility studies, robustness to dilution, globule size analysis, cloud point measurement, transmission electron microscopy and in-vitro dialysis. Transdermal permeation study of nanocarriers and drug suspensions via human skin was performed

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using modified Franz diffusion assembly. SC-SNEDD system was robust to dilution in different media and folds of dilution, maintaining its nano-metric range. SC-nanoemulsion exhibited spherical shaped globules 70 nm in size. Cloud points of all dispersions formed were higher enough than 37°C. In-vitro release from both nanocarriers was significantly higher than drug suspension. Nanoemulsion elaborated could significantly enhance transdermal permeation of SC with higher initial permeation and prolonged release. Paradoxically, SC-SNEDDS exhibited scanty transdermal permeation that could be attributed to low water content of stratum corneum. Nanoemulsion and SNEDDS elaborated exhibited promising in-vitro characteristics for oral sildenafil citrate delivery whereas nanoemulsion elaborated was promising for SC transdermal permeation, as well.

Keywords: sildenafil citrate; nanotechnology; SNEDDS; nanoemulsion; transdermal.

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1 Introduction

Oral therapy of sildenafil citrate (Viagra tablets) is the first line therapy for most men with erectile dysfunction. Parallel to its substantiality in erectile dysfunction therapy, sildenafil citrate (SC) oral delivery suffers numerous drawbacks. These encompass poor bioavailability, considerable first pass effect (70% of the oral dose), numerous side effects, relatively long onset and short duration of action. Furthermore, the drug possesses challenging physicochemical properties for delivery system formulation; including amphoteric nature, pH-dependent characteristics, scanty membrane permeability and poor solubility in both aqueous and oily phases [1–3]. Proposed solutions to circumvent oral delivery obstacles of SC include incorporation into delivery systems with promising in-vitro and biochemical characteristics, either via oral or transdermal delivery. Nevertheless, so far no delivery system approaches for SC problems have been published.

Drug delivery systems help APIs be delivered in an efficient manner rather than altering their chemical nature or biological activity. Lipid-based nanocarriers – when optimised – could have the potential to enhance drug bioavailability, decrease side effects and drug vulnerability to metabolism via both transdermal and oral routes of administration. Among these, nanoemulsions and more recently self-nanoemulsifying drug delivery systems have gained much attention. These nanomedicines could enhance oral bioavailability of drugs vulnerable to first pass effect – as the case of SC – with subsequent reduced dose and side effect. Furthermore, transdermal permeation of certain nanocarriers could successfully circumvent oral delivery obstacles [4–7].

Nanoemulsions are thermodynamically stable, transparent dispersions of oil and water stabilised by an interfacial film of surfactant molecules. Nanoemulsion provides ultra low interfacial tensions and large o/w interfacial areas. Nanoemulsions have a higher solubilisation capacity than simple micellar solutions and their thermodynamic stability offers advantages over unstable dispersions (emulsions and suspensions) because they can be manufactured with little energy input (heat or mixing) and have a long shelf life. The nano-sized droplets leading to enormous interfacial areas associated with nanoemulsions would influence the transport properties of the drug, an important factor in sustained and targeted drug delivery [6–8]. Nanoemulsions were reported to attain oral lymphatic targeting for drugs with extensive first pass metabolism via simulation and stimulation of chylomicron proteins with subsequent enhanced oral bioavailability [6,9]. Furthermore, enhanced bioavailability after transdermal permeation of nanoemulsions has also been reported [10–14].

On the other hand, self-nanoemulsifying drug delivery systems (SNEDDS) or nanoemulsion liquid pre-concentrates comprise isotropic mixtures of oils with surfactants and co-surfactants. These systems spontaneously emulsify when exposed to GIT fluids to form oil in water nanoemulsions with nano-metric droplet size, in the nano-metric range (<200 nm). When optimised, SNEDDS can gather the pharmaceutical privileges of

nanoemulsion with additional peculiarity. SNEDDS can be filled in hard gelatin capsules due to their anhydrous nature enabling its administration as unit dosage form [15–17]. SNEDDS are well recognised to enhance oral bioavailability of drugs vulnerable to pre-systemic clearance and first pass effect**.** Main mechanisms include increasing membrane fluidity to facilitate transcellular absorption, opening tight junction to allow paracellular transport, inhibiting P-glycoprotein and/or cytochrome enzymes by bioactive surfactants, and stimulating chylomicron production by the lipid. Furthermore, the drug can be loaded in the inner-phase of SNEDDS and therefore be protected against enzymatic hydrolysis in the gastrointestinal tract [18–23]. Realising the vulnerability of sildenafil citrate to CYP metabolism in liver, incorporation into SNEDDS for oral delivery deemed intriguing. In addition to oral delivery, Rane and Anderson [24] have proposed that privileges of SNEDDS may also be feasible in topical applications where the matrix constituents act as skin permeation enhancers. However, relevance of SNEDDS in transdermal application has not been investigated so far.

The current work is a part of work series investigating effect on nanomedicine on improving SC characteristics. In the current investigation, two liquid-lipid based nanocarriers, SNEDDS and nanoemulsion, were developed and evaluated to fit both oral and topical application. Both systems included bioactive excipient Cremophor RH40. Another aim was to assess the potential of SNEDDS and nanoemulsion to enhance sildenafil citrate transdermal permeation across human skin for an efficient surrogation of oral delivery.

2 Materials and methods

2.1 Materials

Sildenafil citrate (SC) was obtained from Sun pharmaceuticals (Mumbai, India). Polyoxy 40 hydrogenated castor oil (Cremophor RH40®) and polyoxy 35 castor oil (Cremophor EL®) were obtained from BASF Co. (Germany). Glycerol monolinoleate (Maisine 35-1®), Propylene glycol monocaprylate (Caproyl 90®), isopropyl myristate (IPM), medium chain triglycerides (Labrafac lipophile® WL 1349), PEG-8 caprylic/capric glycerides (Labrasol®), oleoyl acrogol 6-glycerides (Labrafil® M1944CS) and diethylene glycol monoethyl ether (Transcutol HP®) were kindly donated by Gattefosse Co. (Lyon, France). Apricot kernel oil PEG-6 esters (DUB GPE AB) were a kind gift from Stearinerie Dubois Co. (France). Span 85 (HLB 1.8) was obtained from Sigma Chemical Co., St. Louis, MO. Propylene glycol and Tween 80 were obtained from Al-Nasr Pharmaceutical Co. (Egypt). All other chemicals used were of analytical grade.

2.2 Preliminary investigations

Solubility studies of sildenafil citrate in various system components (oils, surfactants and co-surfactants) and buffers were carried out using shake flask method. An excess amount of sildenafil citrate was added into each vehicle followed by vortex mixing for 30 seconds (GEMMY vortex mixer; VM-300, Germany). Mixtures were shaken for 48 h at 30ºC in a thermostatically controlled shaking water bath (Kottermann, type 3047, Hanigsen, Germany), followed by equilibrium for 24 h. Mixtures were then

filtered through a Millipore membrane filter (0.45 μ). Samples were suitably diluted with methanol and drug concentration was obtained by HPLC analysis. Each sample was measured against its blank ingredient (without drug). The experiment was repeated in triplicates. Results were represented as mean value (mg/ml) ± S.E.M.

2.3 Preparation of SC-loaded SNEDDS

Based on the results of preliminary screening, development of sildenafil citrate-loaded SNEDDS utilised one of the optimised placebo SNEDD systems previously formulated by our work group [15]. The system encompassed Maisine 35-1 (16.4%), Caproyl 90 (32.8%), Cremophor RH40 (32.8%) and propylene glycol (16.4%). Briefly, system components (surfactant /co-surfactant mixture with oily phases) were mixed and shaking was proceeded at 100 rpm, 60°C for 30 min in a thermostatically controlled shaking water bath (Kottermann, type 3047, Hanigsen, Germany). Sildenafil citrate was dispersed in the system using Julabo sonicator (USR-3, Ceelbach, Germany). Drug loaded system was then shaken at 100 rpm/60°C for 30 min. Solubility study of sildenafil citrate in the selected system was then carried out. Different drug doses (0.5, 0.6, 0.83 and 1 gm% w/w) were added to the system. The maximum dose yielding clear blend was then selected to prepare sildenafil-loaded SNEDD system.

2.4 Preparation of SC-loaded nanoemulsion

Placebo nanoemulsion pre-concentrate was diluted with water in 1 : 50 folds. Afterwards, sildenafil citrate was added (0.1% w/v) and its dispersion was aided by sonication for 15 minutes. Sildenafil citrate-loaded nanoemulsion was also prepared using carbonate buffer (pH = 10.3) as external phase, to assess the potential of the system to take the peculiarity of higher pH in enhanced drug permeability via biological membranes [25].

2.5 Robustness to dilution

Robustness of sildenafil citrate-loaded SNEDDS to dilution in different volumes and media was assessed. SNEDDS dilution was carried out in four media, namely 0.1 N HCl (pH = 1.2), phosphate buffer (pH = 7.4), carbonate buffer (pH = 10.3) and distilled water. In each medium, three dilution folds (50, 100 and 1000) were compared. Samples were examined for any physical changes (precipitation, separation …) and monitored for 24 hours.

2.6 Globule size analysis

Globule size of sildenafil citrate-loaded nanoemulsion was assessed using laser diffraction particle size analyser (Cilas, model 1064 liquid). Furthermore, the effect of dilution media (type and volume) on SNEDDS globule size was studied. Only robust media to dilution were considered in this study. In each media, three dilution folds (50, 100, and 1000) were compared. Data were represented as mean globular size ± S.D.

2.7 Cloud point measurement

The cloud points of sildenafil citrate-loaded nanoemulsion and SNEDDS diluted (1 : 100) in different media were assessed. Samples diluted in 0.1 N HCl, phosphate buffer (pH = 7.4) and water were subjected to gradual elevation in temperature, starting from 20°C. At cloud point, the drop in original percentage transmittance was measured spectrophotometrically at wavelength of 638.2 nm by UV-160A double beam spectrophotometer (Shimadzu, Japan) [19].

2.8 Transmission electron microscopy (TEM)

Morphological characterisation of sildenafil citrate-loaded nanoemulsion and SNEDDS was carried out using transmission electron microscope (Jeol, JEM-100 CX electron microscope). After proper sample dilution with water, 1–2 sample drops were loaded to copper grid, followed by staining with uranyl acetate solution for 30 s.

2.9 In-vitro drug release

In-vitro drug release was carried out based on dialysis bag method [15]. Sildenafil citrate release from elaborated nanomedicines was compared to its release from aqueous suspension (0.1% w/v) in the same final concentration, using the same sink conditions. Drug suspension in water (7.5 ml), nanoemulsion (7.5 ml), and SNEDDS (1.125 gm) were added to dialysis tubes (Visking® 36/32, 27 mm, MWCO 12000–14000, Serva, USA). Dialysis tubes were then fixed to 100 ml stoppard glass container containing 50 ml citrate buffer (pH = 5). Samples of 3 ml were withdrawn from release medium at fixed intervals (1, 3, 5, 18 and 24 h). Compensation with the same volume fresh citrate buffer was then carried out. Samples – in triplicates – were measured using HPLC analysis. Results were represented as percentage release ± S.E.M.

2.10 Transdermal permeation study

The skin permeation of sildenafil citrate from the elaborated nanomedicines was assessed using modified Franz diffusion assembly [26]. Intact, full thickness human skin obtained from arm plastic surgery was utilised for the study. The subcutaneous fatty tissue was removed from the skin using a scalpel and surgical scissors. The skin surface was cleaned with Ringers' solution and allowed to dry. Skin was completely covered by saline-immersed tissue, packed in aluminium foil and stored in a polyethylene bag at –20°C. Under these conditions, human skin was reported to be stable for 3–6 months [27]. All investigations were conducted in full compliance with ethical principles of Mustafa Elnaggar Hospital (Semoha, Alexandria, Egypt).

The skin samples were mounted on modified Franz diffusion cells with a surface of 3.14 cm^2 and a receptor volume of 8.5 ml. The skin was fixed such that the dermal side of the skin was exposed to the receptor fluid and the hypodermis remained in contact with the donor compartment. The receptor fluid consisted of citrate buffer solution (pH = 5). After an equilibration time of 15 min at 32°C, formulations were applied in the donner compartment in direct contact to the skin. Diffusion cells were placed

in thermostatically controlled shaking water bath (Kottermann, type 3047, Hanigsen, Germany). Diffusion cells were shaken at 100 rpm; while the temperature was maintained at 32°C, simulating skin surface temperature. Sampling was done at 0.5, 1.5, 5, and 24 h. At each point, 3 ml sample was withdrawn and replaced with fresh buffer. All experiments were carried out in triplicate.

2.11 HPLC analysis

A previously validated HPLC system [28] was utilised in the current investigation with minor modifications. In brief, the HPLC instrument (Perkin Elmer series 200, Perkin Elmer instruments, Norwalk, CT 06859, USA) was equipped with reversed-phase C18 column (25 cm × 4.6 mm, particles size = 5 μm). The isocratic mobile phases, 20 mM KH2PO4 (pH 4.7): acetonitrile (25 : 75) was run at a flow rate of 1.0 ml/min at room temperature and the column effluent was monitored by an UV detector set at 293 nm. A volume of 20 μL of each sample was automatically injected into the analytical column. Samples were injected concurrently within the standard solutions injections. The system was also equipped with chromatography interface 600 series link operated by TotalChrom chromatography data system software version 6.2. The calibration curve of peak area against SC concentration was $Y = 862402.4\,(X) - 42324.3$, under SC concentration of 0.5–9 mg%. Retention time was 4.3 ± 0.3 minutes ($R^2 = 0.9956$, LOQ = 0.7 μg/ml, Accuracy 96.6%).

2.12 Statistical analysis

Statistical analysis of the results was carried out using Student's *t*-test ($p < 0.05$).

3 Results and discussion

Preliminary screening of nano-system components was based on solubility studies of sildenafil citrate in various surfactants, co-surfactants and oily phases. Oily phases screened encompassed Maisine 35-1, Isopropyl myristate, Labrafac and Caproyl 90. Surfactants screened included Tween 80, Cremophor RH40, Cremophor EL, and Labrasol. Co-surfactants encompassed Labrafil, Propylene glycol, DUB GPE AB and Transcutol. As demonstrated in Table 1, the ingredients exhibited highest drug solubilisation were Cremophor RH 40 (as surfactant), propylene glycol (as co-surfactant) and Maisine 35-1 (as oily phase). The selected system ingredients constituted matrix components of one of the optimised self-nanoemulsifying systems previously elaborated by our work group (15). The system encompassed Maisine 35-1 (16.4%), Caproyl 90 (32.8%), Cremophor RH40 (32.8%) and propylene glycol (16.4%). Caproyl 90 was involved in the system as Maisine 35-1 alone could not achieve self-nanoemulsifying potential. As demonstrated in ternary phase diagram of the SNEDD system (Figure 1) the system could yield nanoemulsion containing as high as 70% oily phase composition, indicating high emulsification efficiency.

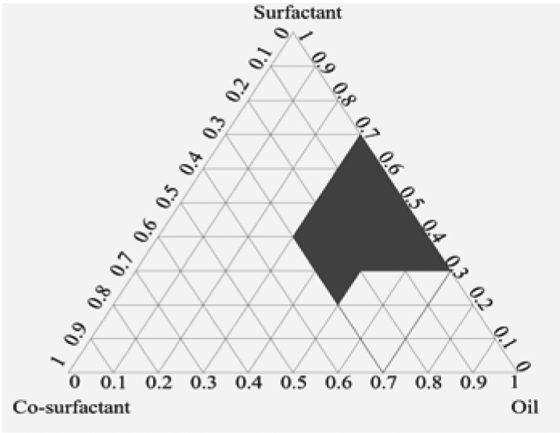
Solubility study of sildenafil citrate in the self-nanoemulsifying drug delivery system designated was carried out in order to attain maximum sildenafil citrate loading. In this context, different drug doses (0.5, 0.6, 0.83 and 1 gm% w/w) were tested for maximum

dose yielding clear mixture. It was found that doses higher than 0.6 g% (40 mg/6 gm formulation) exhibited turbid mixture. Consequently, drug dose of 0.6 g% was selected for preparation of sildenafil citrate loaded SNEDD system.

Table 1 Solubility studies of sildenafil citrate in different surfactants, co-surfactants and oily phases (mean ± S.D.)

Ingredient	*Solubility* (mg/ml)
Surfactants	
Labrasol	2.44 ± 0.02
Cremophor El	8.70 ± 0.57
Cremophor RH40	15.6 ± 0.01
Tween 80	8.8 ± 0.06
Co-surfactants	
Transcutol	5.08 ± 0.44
Propylene glycol	5.66 ± 0.93
DUB GPE AB	0.09 ± 0.01
Labrafil	0.10 ± 0.01
Oily phases	
Caproyl 90	0.56 ± 0.06
Maisine 35-1	3.93 ± 0.93
IPM	0.28 ± 0.01
Labrafac	0.20 ± 0.01

Figure 1 Ternary phase diagram of the self-nanoemulsifying drug delivery system utilised[15]



3.1 Robustness to dilution

The main feature of self-nanoemulsifying drug delivery systems is their ability to spontaneously emulsify under gentle stirring conditions into nanoemulsions. Consequently, a substantial assessment of SNEDD system is its potential to withstand

various dilutions in different physiological pH values. This step is considered crucial for lipid-based systems containing high percentage of soluble surfactants (Type III of lipid formulation classification system LFCS), as the case of SC-SNEDDS prepared in this study [29]. Robustness results contended that all dilutions made in HCl, phosphate buffer (pH = 7.4) and water adopted clear appearance or bluish tang. The dispersions formed exhibited no signs of precipitation, cloudiness or separation for 24 h.

It has been reported that sildenafil citrate possesses high permeability via biological membranes in alkaline pH media in range of 8–11 [25]. Consequently, placebo SNEDD system dilution in carbonate buffer (pH 10.3) was carried out to assess this buffer as nanoemulsion external phase in comparison to water. It is noteworthy that concentrated nanoemulsion (50 and 100 folds dilution) is required to attain reasonable final drug loading. Up on dilution of placebo SNEDD system in alkaline buffer, crude emulsions were formed with 50 and 100 fold dilutions, reflecting loss of nano-emulsifying property. For confirmation, sildenafil citrate loaded nanoemulsion was reformulated with carbonate buffer as external phase. Crude emulsion was formed instead of nanoemulsion. Consequently, carbonate buffer was excluded as external phase for sildenafil citrate loaded nanoemulsion.

3.2 Globule size analysis

Globule size of nanocarriers is an imperative parameter influencing their intended performance. Improved drug bioavailability via different administration routes after incorporation into nanoemulsion is based crucially on the nano-metric globular size (< 200 nm) (6–10). In this work, globule size analysis in the media where SNEDD system exhibited robustness to dilution was carried out. Consequently, globule size analysis of nanoemulsions formed in 0.1 N HCl, phosphate buffer (pH 7.4) and water each in three dilutions (50, 100 and 1000 folds) was assessed. Results are depicted in Table 2. All dispersions formed were in the nano-range. As for nanoemulsion, a globular size of 70 nm was exhibited. Results of globular size analysis reflect the capability of the elaborated SNEDD system of sildenafil citrate to withstand gradual dilution in GIT fluids.

Table 2 Characterisation of sildenafil citrate-loaded SNEDDS elaborated

Characteristics	*Water*	*0.1 N HCl*	*Phosphate buffer* (*pH 7.4*)
Globule size (nm)			
(50 folds)	140 ± 8.25	130 ± 7.00	140 ± 20.32
(100 folds)	20 ± 1.40	22 ± 1.98	20 ± 2.12
(1000 folds)	10 ± 1.72	10 ± 0.98	10 ± 1.74
Cloud point (ºC)	65	73	70

3.3 Cloud point measurements

The cloud point is the temperature above which the formulation clarity turns into cloudiness [19]. At higher temperature, phase separation can occur due to dehydration of polyethylene oxide moiety of the non-ionic surfactant, affecting both drug solubilisation and formulation stability. The cloud point of the formulation should therefore exceed

37°C numerous factors affecting cloud point are reported elsewhere. Cloud point of SC loaded SNEDD system elaborated was assessed in different pH media (water, 0.1 N HCl, phosphate buffer pH 7.4). As depicted in Table 2, cloud points of nanoemulsions formed in all media were sufficiently higher than 37°C. SNEDD system diluted in water exhibited cloudiness at 65°C, with a drop in % transmittance from 92% to 40%. Dilution in 0.1 N HCl possessed cloud point of 73°C with % transmittance reduction from 86% to 46%. Phosphate buffer dilution, on the other hand, turned cloud at 73°C with transmittance drop from 88% to 50%. As for sildenafil citrate loaded nanoemulsion, cloudiness was detected at 80°C. In all formulations, cloudiness was reversible after minutes. Results contented the stability of sildenafil citrate loaded nanoemulsion and SNEDD system towards separation in the GIT temperature and pH.

3.4 Transmission electron microscopy

Morphological examination of sildenafil citrate loaded SNEDDS and nanoemulsion was performed using transmission electron microscope (TEM). Sildenafil citrate loaded nanoemulsion and SNEDDS samples were properly diluted with distilled water (1 : 1000). As depicted in Figures 2 and 3, nanoemulsion and SNEDD system (respectively) have demonstrated spherical globules with regular shaped morphology.

Figure 2 TEM photograph of sildenafil loaded nanoemulsion with 1000 folds dilution scale bar is 1 µm

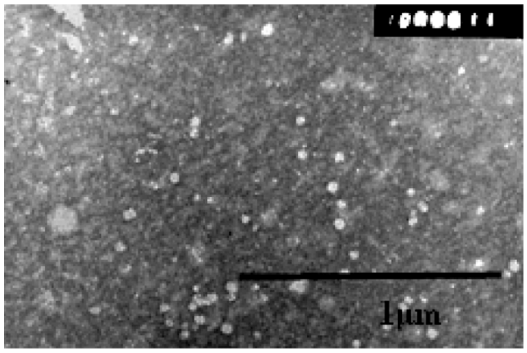
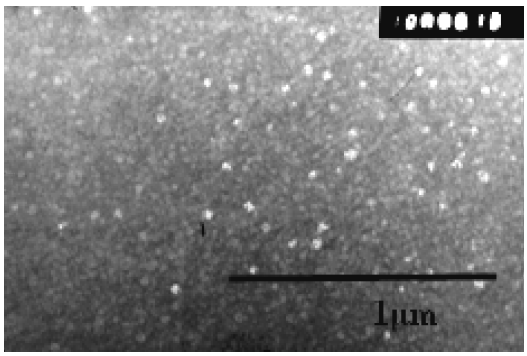


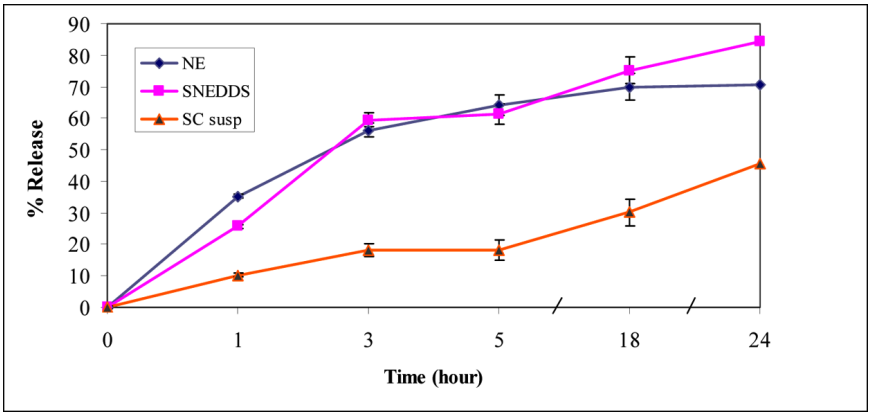
Figure 3 TEM photograph of sildenafil citrate SNEDDS with 1000 folds dilution scale bar is 1 µm



3.5 In-vitro drug release

In-vitro release of sildenafil citrate from SNEDDS and nanoemulsion was assessed using dialysis bags with proper MWCO value (12–14,000) for secure separation of drug molecules from delivery system components. Solubility study of SC in different buffer systems including citrate buffer (pH = 5), phosphate buffer (pH = 7.4) and carbonate buffer (pH = 10.3) was carried out. It was found that drug solubility was significantly decreasing with pH increase. Drug solubility was lowest in carbonate buffer (0.05 mg/ml) followed by phosphate buffer (8.58 mg/ml) and was highest with citrate buffer (2.25 mg/ml). This could be explained regarding the basic functional group of the drug (NH-piperazine, pKa 8.7). The reverse relationship between sildenafil solubility and solvent pH is reported elsewhere [1]. Consequently, drug release testing was carried out in citrate buffer to attain proper sink condition. Drug release from the two nanocarriers was compared to that from drug suspension in the nanoemulsion external phase (water). The applied sample volume has considered unified amount of drug applied inside dialysis bag (7.5 mg) for all samples. This would consequently circumvent the influence of concentration gradient on the release rate. As depicted in Figure 4, drug release from both nanocarriers was significantly higher than drug suspension. Sildenafil citrate loaded nanoemulsion exhibited higher initial release (in the first three hours of the experiment) followed by sustained release compared to SNEDDS. Slower initial release from SNEDDS could be attributed to the time consumed in self-nanoemulsification of the nanoemulsion pre-concentrate.

Figure 4 In-vitro release profiles of sildenafil citrate from SNEDDS and nanoemulsion compared to drug suspension in citrate buffer (pH = 5) (see online version for colours)

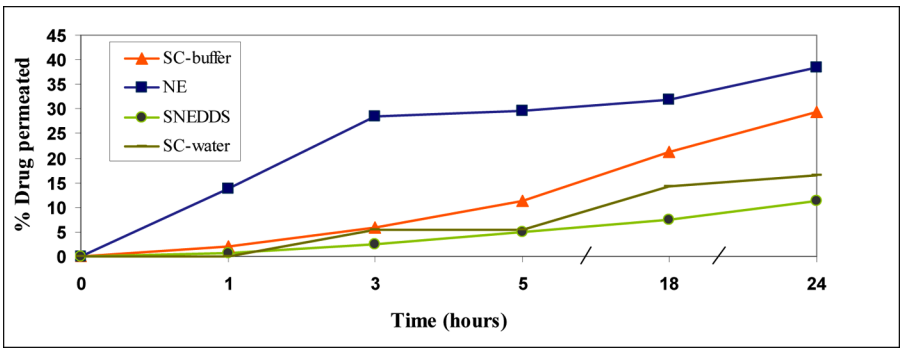


3.6 Transdermal permeation studies

Transdermal permeation of SC from the two nanocarriers elaborated was compared to that from drug suspensions in two solvents (water and carbonate buffer pH = 10.3) to detect the effect of pH independently on human skin permeation of SC and compare to delivery system effect. Furthermore, each nanocarrier sample was compared to its placebo formulation and each drug suspension was compared to its placebo solvent for comparison. The applied sample volume has considered unified amount of drug in all formulations (0.75 mg). As depicted in Figure 5, drug permeation rate from

nanoemulsion was significantly higher than both drug suspensions. Furthermore, nanoemulsion exhibited higher initial drug permeation followed by sustained release compared to drug suspensions, a promising result for shorter onset and longer duration of drug action.

Figure 5 Transdermal permeation profiles of SC loaded nanoemulsion (NE) and SNEDDS compared to drug suspensions in carbonate buffer (pH = 10.3) and water (see online version for colours)



The three main factors generally determining the transdermal efficacy of drugs constitute mobility of drug in the vehicle, release of drug from the vehicle, and permeation of drug into the skin. These factors affect either the thermodynamic activity that delivers the drug into the skin or the permeability of the drug in the skin, particularly the stratum corneum. The small particle size is implicated as large surface area and film formation that consequently creates intimate drug contact with the corneocytes, thus increasing the amount of encapsulated drug/nutrient penetrating into the viable skin [4]. This approach may partially explain the enhanced permeation of sildenafil citrate from nanoemulsion (70 nm). Furthermore, surfactants, which can loosen or fluidise the lipid bilayers of the stratum corneum, can act as permeation enhancers. Compared to microemulsions, transdermal nanoemulsions have been shown to increase bioavailability and efficacy of a number of compounds such as anti-inflammatory agents, dicumarol, and progesterone [13,14,27–30].

Albeit sildenafil citrate exhibited higher release from carbonate buffer suspension (pH = 10.3) compared to water suspension, drug release from nanoemulsion (water as external phase) was higher than lipid nanoparticles that with carbonate buffer as external phase (pH = 10.3) (unpublished data). Theses results propose that formula pH is an intervening but not the sole factor controlling sildenafil citrate permeation via human skin. Other intervening factors exist, that might include carrier particle size and fluidity. Carbonate buffer was not utilised as nanoemulsion external phase as the system was not robust to dilution in this pH (10.3) in the designated dilution fold (1 : 5).

As a paradoxical effect, SNEDDS exhibited poor transdermal permeation of SC via human skin (Figure 5), despite its promising in-vitro dialysis profile and higher concentration of non-ionic surfactant as a permeation enhancer. This effect could be explained considering low water content (20%) of the stratum corneum [31] that is supposed to constitute a handicap against spontaneous nanoemulsification of the self-nanoemulsifying system. Realising that SNEDD system developed encompassed significantly higher surfactant and oil concentrations compared to

nanoemulsion, the priority of particle size over additive effect as factors influencing transdermal SC permeation might be proposed. In spite of its scanty transdermal potential, sildenafil citrate loaded SNEDD system prepared in this work is anticipated to improve oral delivery of SC regarding promising in-vitro characteristics and incorporation of bioactive surfactant, Cremophor RH40, that possesses reported inhibitory effect on cytochrome enzymes responsible for sildenafil extensive hepatic metabolism [32].

4 Conclusions

In the current investigation, liquid lipid-based nanocarriers of sildenafil citrate – nanoemulsion and SNEDDS – have been developed and in-vitro assessed. The delivery systems prepared are characterised by nanometric spherical globules, robustness to different media and folds of dilution, adequately high cloud points and significant improvement in drug release compared to drug suspension. Nanomedicines developed in this paper possesses promising characteristics to improve sildenafil citrate therapeutic performance via oral route. Furthermore, nanoemulsion has successfully enhanced sildenafil citrate transdermal permeation via human skin. Transdermal permeation of SNEDDS via human skin has been proved to be minimum despite enhanced in-vitro release and high content of permeation enhancers.

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