EUROPEAN JOURNAL OF BIOMEDICAL AND PHARMACEUTICAL SCIENCES
http://www.ejbps.com

ISSN 2349-8870
Volume: 3
Issue: 7
522-526
Year: 2016

STUDY ON THE INFLUENCE OF COSOLVENT AND SURFACTANT ON SOLUBILIZATION OF EFAVIRENZ

Mbah Chika, Omeje Edwin* and Ugodi Gerald.

Department of Pharmaceutical and Medicinal Chemistry, Faculty of Pharmaceutical Sciences, University of Nigeria, Nsukka, Enugu State, Nigeria.

***Author for Correspondence: Omeje Edwin**
Department of Pharmaceutical and Medicinal Chemistry, Faculty of Pharmaceutical Sciences, University of Nigeria, Nsukka, Enugu State, Nigeria.,

Article Received on 22/05/2016 **Article Revised on 13/06/2016** **Article Accepted on 02/07/2016**

ABSTRACT
The very poor aqueous solubility of drugs, give rise to difficulties in the design of pharmaceutical formulations and very often lead to variable oral bioavailability. Cosolvency and micellar solubilization are widely used alternative for the solubilization of poorly soluble drugs. Efavirenz has very poor aqueous solubility and belongs to Class II drugs under Biopharmaceutical Classification Systems. The purpose of this work was to investigate the effect of some solubilizing agents on the aqueous solubility of efavirenz and invariably its bioavailability. Determination of aqueous solubility of efavirenz was in aqueous mixture of cosolvents (glycerol, propylene glycol, polyethylene glycol 400) and surfactants (tween 20, tween 80, sodium lauryl sulfate, cetrimide). Excess of efavirenz was added into each of the mixed solvent at 25 ±1^{o}C. Equilibrium method (shake-flask technique) was utilized. Following equilibrium, the solutions were filtered and analyzed using ultraviolet absorption spectroscopic technique. The results indicate logarithm linear relationships between efavirenz solubility in water-cosolvent mixtures and cosolvent fraction volume. The solubilization power (σ) or the slope is used to describe the drug-cosolvent-water system. Linear relationships between the solubility of efavirenz in water-surfactant mixtures and surfactant concentration were observed. The solubilization capacity (k) or the slope is used to describe the drug-surfactant-water system. The free energy of solubilization was found to be linearly related to micellar partition coefficients of efavirenz. The results of the study show that cosolvency and micellization significantly improved aqueous solubility of efavirenz.

KEYWORDS: Efavirenz, aqueous solubility, cosolvency, micellization.

1.1 INTRODUCTION
Most drugs in clinical practice are lipophillic in nature, and this property has contributed to their low aqueous solubility and bioavailability. A number of attempts has been made to develop a system which could increase aqueous solubility, bioavailability and hence reduce the drug dose. Such attempts include but not limited to techniques such as chemical modification, cosolvency, cyclodextrin complexation, micellization, microemulsion, micronization, nanoemulsion, nanoparticles, nanosuspensions, self-emulsifying systems and solid dispersion.[1,2, 3, 4] In the present work, cosolvency and micellization techniques were studied to ascertain the extent the aqueous solubility of efavirenz would be enhanced. Cosolvency (mixing a permissible non-toxic organic solvent with water) is the most common and feasible technique to enhance the aqueous solubility of drugs. Cosolvents have small hydrocarbon regions that are nonpolar and cannot interact strongly with water. They therefore tend to reduce the ability of the aqueous system to squeeze out nonpolar solutes. Typical examples of cosolvents are ethanol (EtOH), glycerol, propylene glycol (PG), polyethylene glycol (PEG400) and tetrahydrofurfuryl alcohol polyethyleneglycol ether (glycofurol).[5, 6, 7] Micellization have been reported to solubilize a number of poorly water soluble drugs.[8, 9,10, 11] Surfactants (amphiphilic molecules composed of a hydrophilic moiety known as the head and a hydrophobic moiety known as the tail) do that by forming micelles (colloidal clusters) in solutions. As association colloids, micelles are formed spontaneously under certain conditions (self assembling system) and are thermodynamically more stable towards both dissociation and aggregation. Depending upon the drug hydrophobicity, it can be solubilized in the inner core of the micelle, on the surface of the micelle or at an intermediate location in the palisade layer. Efavirenz, chemically defined as (4*S*)-6-chloro-4-(cyclopropylethynyl)-1,4-dihydro-4-(trifluoromethyl)-2*H*-3,1-benzoxazin-2-one has chemical structure presented in Figure 1.

The drug is a non-nucleoside reverse transcriptase inhibitor (NNRTI) and structurally belongs to the class of organic compounds known as benzoxazines. Clinically, it is used to treat HIV-1 infection. Its mechanism of action involves direct binding to reverse transcriptase (RT) thereby blocking the RNA-dependent and DNA-dependent DNA polymerase activities resulting in the disruption of the enzyme's catalytic site.[12] Efavirenz is practically insoluble in water, has logarithm partition coefficient (octanol-water) of 4.6. The pharmacokinetics of efaviren shows the drug to have average bioavailability of 40 %, above 90 % protein binding and very high half-life (40-55 h) following administration of a single dose.[13] Efavirenz is available as tablets and capsules respectively. However, the possibility for the development of liquid and parenteral formulations of efavirenz exists if its aqueous solubility is significantly enhanced. Due to the drug's high daily dose (600 mg/day) and low bioavailability leading to sometimes serious side effects such as insomnia, loss of memory and suicidal tendencies, a number of studies have been reported on the enhancement of aqueous solubility and bioavailability of the drug. Such enhancement techniques include solid inclusion complexes[14], solid self emulsifying drug delivery systems[15], solid dispersion[16], inclusion complexation and liquid anti-solvent precipitation.[17] Despite all the various solid alteration techniques that have been used to enhance the aqueous solubility of poorly water soluble drugs, solvent alteration has been shown to be the most effective means of producing a thermodynamically stable increase in aqueous solubility.[7] Literature search has revealed that most of the techniques employed to enhance the aqueous solubility of efavirenz involved solute alteration with little or no information on solvent alteration. Therefore, in the present investigation, solvent alteration was chosen to enhance the aqueous solubility of efavirenz using the cosolvency and micellization techniques.

1.2 MATERIALS AND METHODS

1. 2. 1. Materials

Efavirenz (Mylan Pharmaceuticals, USA), glycerol, propylene glycol, polyethylene glycol 400 (PEG 400), polysorbate 20 (tween 20), polysorbate 80 (tween 80), sodium lauryl sulphate and cetrimide were purchased from Sigma-Aldrich, USA. Methanol (Fisher Scientific, USA) and other chemicals were of analytical reagent grade.

1. 2. 2. Solubility Determination

Excess of drug (50 mg) was placed in a 10 ml sealed vial containing 5 ml of purified water, different concentrations of cosolvents and surfactants respectively. The vials were maintained at 25°C and the contents were shaken on Rotary Flask Shaker for 48 h to achieve equilibrium. The solution was centrifuged and the supernatant was filtered through 0.45 µm filter. The absorbance of the clear solution was determined at λ_{max} of the drug (245 nm) after suitable dilution with the appropriate solvent. The concentration of drug was determined from the calibration graph. The solubility was calculated by multiplying the drug concentration, so obtained, by the appropriate dilution factor. The reported data are an average of three determinations.

1.3 RESULTS AND DISCUSSION

The results of the solubility study of efavirenz in cosolvent and surfactant concentrations are given in Tables 1 and 2 respectively. They are also presented in Figures 2 and Figures 3 respectively. The results in Table 1 show that the solubility of efavirenz increases as the concentration of the cosolvent is increased. Of the cosolvents investigated, the least polar cosolvent, polyethylene glycol 400 produced the highest aqueous solubility of efavirenz and the most polar cosolvent, glycerol gave the lowest aqueous solubility of the drug. For instance, at the highest concentration studied (25% w/v), polyethylene glycol 400 gave a 16-fold increase in the aqueous solubility of efavirenz. The difference in the aqueous solubility of efavirenz in the cosolvents could be due reduction in the ability of the aqueous system to squeeze out nonpolar solutes by the small nonpolar hydrocarbon region in the cosolvent. Plots in Figure 2, depict that the greater the difference in the polarity of the solvents in a given mixed solvent, the greater was the solubilization power (α).

The values of the solubilization power are given in Table 1. Figure 2 also shows exponential increase in efavirenz aqueous solubility with the concentrations of the cosolvents. The logarithmic relationship between total drug solubility (S_{tot}) in a mixed solvent and cosolvent concentration (C_{cosol}) can be described by Equation 1.[5,7]

$$\log S_{tot} = \log S_w + \sigma C_{colsol} \quad (1)$$

where S_{tot} is total drug solubility, S_w is drug solubility in water and σ is cosolvent solubilization power.

The influence of surfactants on the aqueous solubility of efavirenz, are given in Table 2 and Figure 3 respectively. From Table 2, it would be seen that sodium lauryl sulfate out of the surfactants investigated, gave the greatest enhancement in the aqueous solubility of efavirenz. At the highest concentration studied (2% w/v), a 23-fold increase in the aqueous solubility of the drug was observed. The increase could arise from the micellar properties and not the alkaline properties of the surfactant as efavirenz has a high pKa value. In Figure 3, a linear relationship between the total solubility of the drug in water-surfactant mixtures and surfactant concentration was observed. The slightly higher *k* value of tween 80 when compared to tween 20 may be the result of its longer alkyl chains, therefore, indicating that efavirenz might have been solubilized in the hydrophobic core of the micelles. Equation 2 describes the relationship between efavirenz solubility in a micellar solution and surfactant concentration.[18]

$$S_{tot} = S_w + kC_{surf} \quad (2)$$

where C_{surf} is the concentration of micellar surfactant (*i.e.*, the total surfactant concentration minus the critical micellar concentration) and *k* is the molar solubilization capacity (the number of moles of solute that can be solubilized by 1 mole of micellar surfactant). The equation shows that the critical micellar concentration (cmc) is much lower than C_{surf}. The molar solubilization capacity *k* is obtained from the slope of a plot of the total solubility of the drug versus surfactant concentration or by using Equation 3.

$$k = S_{tot} - S_w/C_{surf} \quad (3)$$

Another descriptor that defines micellar solubilization of efavirenz is the micelle-water partition coefficient and is defined as the ratio of the amount of drug in water to the amount of drug in micelle. Mathematically, it is expressed as:

$$P = S_{tot} - S_w/S_w \quad (4)$$

In this study, equation (4) was used to calculate the partition coefficient of efavirenz in the micellar solutions.

As cosolvent and surfactant solubilization are thermodynamic processes, the standard free energy (ΔG^o) involved in efavirenz solubilization was calculated using Equation 5[19] for cosolvent.

$$\Delta G^o = -2.303RT \log S_{cosol}/S_{water} \quad (5)$$

where S_{cosol} is drug solubility in cosolvent; S_w is drug solubility in water; R is gas constant; T is the absolute temperature and Equation 6[20] for micellar solubilization.

$$\Delta G^o = -RT \ln P \quad (6)$$

The spontaneity of the solubilization process is shown by the negative values obtained for the standard free energy (Table 3). Polyethylene glycol 400 when compared to other cosolvents, has the greatest tendency of solubilizing efavirenz as indicated by its largest negative free energy value at the maximum concentration (25 % w/v) investigated. Similarly, at surfactant maximum concentration (2 % w/v), sodium lauryl sulfate exhibited the largest negative free energy value. To correlate the two parameters, the micellar partition coefficient was plotted against the standard free energy (ignoring the negative sign). A linear relationship was observed in all the plots with the correlation coefficient (R^2) of 0.9646, 0.9221, 0.9279 and 0.9821 for tween 20, tween 80, sodium lauryl sulfate and cetrimide respectively. Figure 4, illustrates the plot of standard free energy versus micellar partition coefficient (R^2 = 0.9279) for sodium lauryl sulfate.

Table 1: Solubilization of efavirenz by cosolvents and solubilization parameters.

Cosolvent Conc. (% w/v)	Solubility (ug/ml)			Solubilization Parameter, σ ($\%^{-1}$)		
	Glycerin	Propylene Glycol	Polyethylene Glycol 400	Glycerin	Propylene Glycol	Polyethylene Glycol 400
0.0	8.667±0.064	8.667±0.064	8.667±0.064	0.0241	0.0296	0.0440
5.0	11.999 ±0.487	13.483 ±0.248	17.151 ±0.088			
10.0	13.776 ±0.315	17.998 ±0.064	31.769 ±0.256			
15.0	17.109 ±0.068	27.133 ±0.073	47.326 ±0.188			
20.0	22.377 ±0.196	36.644 ±0.272	78.524±0.268			
25.0	32.166 ±0.098	51.844 ±0.184	137.309±0.182			

Table 2. Solubilization of efavirenz by surfactants and solubilization parameters.

Surfactant Conc. (% w/v)	Solubility (μgml^{-1})				Solubilization Parameter, *k* ($\mu gml^{-1}/\%wtv^{-1}$)			
	Tween 20	Tween 80	Sodium lauryl sulfate	Cetrimide	Tween 20	Tween 80	Sodium lauryl sulfate	Cetrimide
0.00	8.667±0.064	8.667 ±0.064	8.667±0.064	8.667±0.064	18.4	35.4	89.5	11.2
0.10	14.443±0.252	20.776±0.098	39.442±0.148	11.999 ±0.105				
0.20	20.443±0..212	31.331±0.165	63.773±0.124	14.887 ±0.112				
0.40	26.553±0..093	40.442±0.150	85.551±0.272	18.443±0.152				
0.50	32.442±0.205	51.107±0.069	110.771±0.152	22.331±0.402				
1.00	38.442±0.076	63.329±0.186	150.991±0.072	26.553±0.270				
2.00	48.552±0.037	85.770±0.099	200.871±0.398	32.442±0.214				

Table 3: Standard free energy involved in solubilization of efavirenz

Cosolvent Conc. (% w/v)	Δ G° (J/mol)			Surfactant Conc. (% w/v)	Δ G° (J/mol)			
	Glycerin glycol	Propylene glycol 400	Polyethylene		Tween 20	Tween 80	Sodium lauryl sulfate	Cetrimide
0.0	-	-	-	0.00	-	-	-	-
5.0	-806.1	-1095.2	-1691.5	0.10	1005.7	-828.8	-3140.2	2369.2
10.0	-1148.5	-1810.9	-3219.0	0.20	-759.6	-2382.2	-4584.0	822.0
15.0	-1685.4	-2828.2	-4206.8	0.40	-1795.5	-3219.5	-5409.3	-298.3
20.0	-2350.6	-3572.8	-5461.6	0.50	-2500.7	-3936.7	-6112.3	-1128.2
25.0	-3249.9	-4432.8	-6846.5	1.00	-3058.4	-4563.9	-6935.4	-1795.5
				2.00	-3782.8	-5416.3	-7679.9	-2501.7

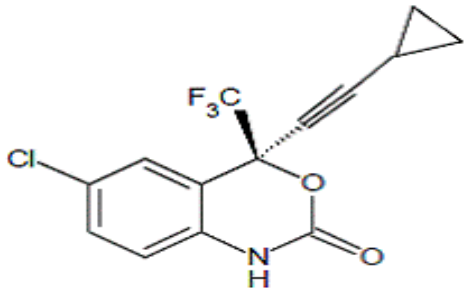

Figure 1: Chemical structure of efavirenz.

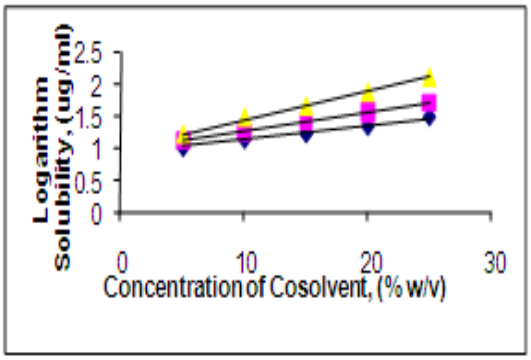

Figure 2: Plot of logarithm solubility (µg / ml) of efavirenz versus cosolvent concentration (% w/v)

□-------□ Glycerol
□-------□ Propylene glycol
Δ-----Δ Polyethylene glycol 400

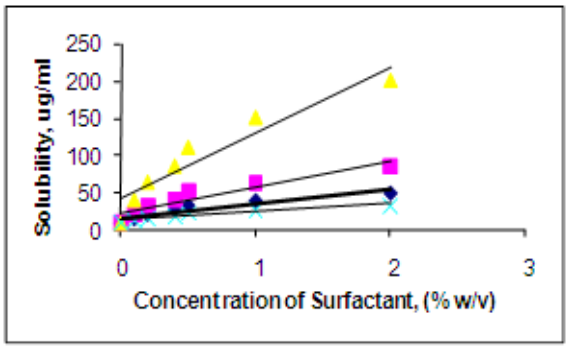

Figure 3: Plot of aqueous solubility (µg/ml) of efavirenz versus surfactant concentration (% w/v)

×-----× Cetrimide

□-------□ Tween 20
□-------□ Tween 80
Δ--------Δ Sodium lauryl sulfate

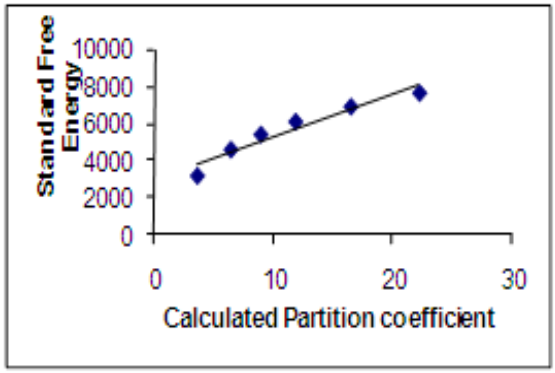

Figure 4: Plot of standard free energy of solubilizing efavirenz in sodium lauryl sulfate solution versus micellar partition coefficient.

4. CONCLUSION

The vehicles used in this study are generally considered safe for oral administration at the concentration levels investigated.[21,22] At the maximum concentration studied for cosolvency (25% w/v) and micellization (2% w/v) respectively, efavirenz solubility is observed to be significantly enhanced. However, the results obtained demonstrate that aqueous solubility is still a limiting factor for efavirenz bioavailability as literature and official reports have stated that for a drug to be considered highly soluble, the dose: solubility ratio should be less than 250 Ml.[23,24, 25] The study also found that a linear relationship exists between standard free energy and the micellar partition coefficient of the drug. This implies that small free energy is required for biological membrane permeation of formulation in which efavirenz has high micellar partition coefficient. Cosolvent or surfactant solubilization parameters such as solubilization power or molar solubilization capacity, micelle–water partition coefficient, and standard free energy of solubilization gave quantitative estimate of the solubilization efficiency of the cosolvent or surfactant system investigated. Of the vehicles studied, none was found to be able to give the minimum drug solubility that

is required for 5-ml or 2-ml dose of the smallest unit dose (50 mg) of the drug. The result, therefore, suggests that oral liquid or parenteral formulations of efavirenz at the highest concentration investigated are not applicable, but some might be incorporated into solid dosage forms (tablets or capsules) as excipients.

REFERENCES

1. Shafiq S, Shakeel F, Talegaonkar S, Ahmed FJ, Khar RKM, Ali M. Development and bioavailability assessment of ramipril nanoemulsion formulation. Eur J Pharm Biopharm., 2007; 66: 227-43.
2. Lee PJ, Lenger R, Shestri VP. Novel microemulsion enhancer formulation for simultaneous transdermal delivery of hydrophilic and hydrophobic drugs. Pharm Res., 2003; 20: 264-9.
3. Mbah CJ. Solubilization of valsartan by aqueous glycerol, polyethylene glycol and micellar solutions. Die Pharmazie., 2006; 6: 322-4.
4. Li P, Zhao LW, Yalkowsky SH. Combined effect of cosolvent and cyclodextrin on solubilization of non-polar drugs. J Pharm Sci., 1999; 88: 1107-11.
5. Ran Y, Zhao L, Xu Q, Yalkowsky H. Solubilization of cyc-losporin A. AAPS Pharm Sci Tech., 2001; 2(1): 2.
6. Yalkowsky SH, Roseman TJ. Solubilization of drugs by co-solvents. In: Yalkowsky SH, ed. Techniques of Solubilization of Drugs. Vol 12. New York, NY: Marcel Dekker Inc., 1981; 91-134.
7. Zhao L, Li P, Yalkowsky SH. Solubilization of fluasterone. J Pharm Sci., 1999; 88: 967-9.
8. Krishn AK, Flanagan D R. Micellar Solubilization of a new antimalarial drug, β-arteether. J. Pharm. Sci., 1989; 78: 574-6.
9. Alkhamis K A, Allaboun H, AL-Mornani WY. Study of the solubilization of gliclazide by aqueous micellar solutions. J Pharm Sci., 2003; 92: 839-46.
10. Seedher N, Kanojia M. Micellar solubilization of some poorly soluble antidiabetic drugs. AAPS Pharm. Sci Tech., 2008; 9: 431-6.
11. Liu C., Desai K. G. H., Liu C. Solubility of valdecoxib in the presence of ethanol and sodium lauryl sulfate at (298.15, 303.15 and 308.15K) J. Chem. Eng. Data., 2004; 49: 1847–1850.
12. Physcian Desk Reference. 59th Edition, Thomson PDR, New Jersey, 2005; 2216.
13. Sosnik A, Carcaboso AM. Drug delivery systems in HIV pharmacotherapy: What has been done and the challenges standing ahead. J Control Rel., 2009; 138: 2-15.
14. Chaitanya V, Narayan PN, Kumar S, Chowdary KPR. (2014). Enhancement of solubility dissolution rate and formulation development of efavirenz tablets employing β-cd and lutrol: a factorial study. J Global Trends Pharm Sci., 2014; 5: 1356-60
15. Reddy MS, Reddy NS, Reddy SM. Solubility Enhancement of Poorly Water Soluble Drug Efavirenz by Solid Self Emulsifying Drug Delivery Systems. Int J Pharm Res & Rev., 2014; 3(4): 20-8.
16. Chowdary KRK, Enturi V. Enhancement of dissolution rate and formulation development of efavirenz tablets employing starch citrate-a new modified starch. J Appl Pharm Sci., 2011; 5: 119-123.
17. Mahapatra1 AK, Murthy PN. Solubility and dissolution rate enhancement of efavirenz by inclusion complexation and liquid anti-solvent precipitation technique. J Chem Pharm Res., 2014; 6(4): 1099-1106.
18. Ong JTH, Manoukian E. Micellar solubilization of timbesone acetate in aqueous and aqueous propylene glycol solutions of nonionic surfactants. Pharm Res., 1988; 3: 704-8.
19. Feldman SL, Gibaldi M. Effect of urea on solubility. Role of water structure. J Pharm Sci., 1967; 56: 370-5.
20. Rangel-Yagui CO, Junior AP, Tavares C. Micellar solubilization of drugs. J Pharm Sci., 2005; 8: 147-63.
21. Mottu F, Laurent A, Rufenacht D, Doelker E. Organic solvents for pharmaceutical parenterals and embolic liquids: A Review of toxicity data. PDA J Pharm Sci Tech., 2000; 54: 456-69.
22. Sweetana S, Akers MJ. Solubility principles and practices for parenteral drug dosage form development. PDA J Pharm Sci Tech., 1996; 50: 50-6.
23. Lindenberng M, Kopp S, Dressman JB. Classification of orally administered drugs on the World Health Organization model list of essential medicines according to the biopharmaceutics classification system. Eur J Pharm Biopharm., 2004; 58: 265-78.
24. Strauch S, Jantratid E, Dressman J.B, Junginger HE, Kopp S, Midha KK, Shah VP, Stavchansky S, Barends DM. Biowaiver monographs for immediate release solid oral dosage forms: lamivudine. J Pharm Sci., 2011; 100: 2054-63.
25. United States. Department of Health & Human Services. Food and Drug Administration. Center for Drug Evaluation and Research. *Guidance for industry*: waiver of *in vivo* bioavailability and bioequivalence studies for immediate-release solid oral dosage forms based on a biopharmaceutics classification system. Rockville: FDA, 2000; 1-13.