Influence of B-Cyclodextrin as An Inclusion Complexing Agent For The Solubility of Mefenamic Acid in Base Solution

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Mefenamic acid is a poorly water soluble non steroidal anti-inflammatory drug (NSAID). The solubility of mefenamic acid can be increased by forming inclusion complexes with β-cyclodextrin. Inclusion complexes can be performed when the drug molecules have a suitable size with β-cyclodextrin and it is influenced by the ionized/unionized form of drug molecules. The purpose of this study is to determine the effect of β-cyclodextrin for enhancing the solubility of mefenamic acid in base solution (pH 7,0: 8,0: 9,0: 10,0). This study is also considering the pKa value of mefenamic acid (pKa = 4.2) because it forms complexes with β-cyclodextrin in base solution. It is expected with the increasing of pH value, there will be more ionized molecule so it could influenced the performing of inclusion complexes. Phosphate buffer was used to make base solution of pH 7.0 and pH 8.0; and borate buffer was used for pH 9.0 and 10.0. Procedure to prepare the inclusion complexes was stirring or shaking β-cyclodextrin $2x10^{-3}$M solution with excessive mefenamic acid at temperature of 37±0.5^{o}C. A supersaturated solution of mefenamic acid was formed after shaking the sample in waterbath shaker for 5 hours. Detection of inclusion complexes in solution using UV-Vis spectroscopy. The results showed that there were solubility increases in pH 7.0 and pH 8.0 27.07% and 26.57%, respectively. But there were no solubility increases showed in pH 9.0 and pH 10.0 because there were plenty of ionized mefenamic acid. It showed that pKa value affected the performing of inclusion complexes of mefenamic acid-β-cyclodextrin in base solution.

Keywords : *mefenamic acid; β-cyclodextrin; inclusion complex; base solution; solubility*

INTRODUCTION

Mefenamic acid is a non steroidal anti-inflammatory drug (NSAID) effective in treating fever, pain, and inflammation in the body. The chemical name is *N-(2,3 xylil) anthranilic acid* and its molecule formula is $C_{15}H_{15}NO_2$ (Fig. 1) (Depkes RI, 1995). It is white to grayish white microcrystalline powder, odourless, soluble in solution of alkali hydroxides, sparingly soluble in chloroform and practically insoluble in water (Budavari S., 2001). It has pKa value of 4.2. The aqueous solubility of mefenamic acid at pH 7.1 is 0.0041g/100 ml (25^0C) and 0.008g/100 ml (37^0C) (Bekers, *et. al*.,). The solubility of mefenamic acid can be altered by addition of β-cyclodextrin.

β-cyclodextrin is cyclic oligosaccharides composed of dextrose units join through a 1,4 bond with seven dextrose units (Fig. 2). The shape of these molecule is similar to a truncated cone which has a hydrophilic outer surface and a hydrophobic inner cavity (Great Britain The Government on Health, 1993). It is white crystalline powder, odourless and has a little sweet taste (Raymond , *et al*., 2003). Many insoluble drugs can enhance their solubility and bioavalability after formed inclusion complexes with β-cyclodextrin, such as : Acyclovir (Rossel , et al., 2000); tolnaftate (Peri , et al., 1994); celecoxib (Reddy , 2004); ketoprofen (Lu, et al., 2004); and nimesulide (Nalluri, et al., 2003). From three types of cyclodextrin, β-cyclodextrin has the lowest solubility in water than others because there are so many intramolecular hydrogen bond between secondary hydroxyl components that prevent hidration from β-cyclodextrin molecules (Bogdan , *et al.*, 2006).

Inclusion complexes are formed if drug molecules are included in the β-cyclodextrin cavity. The minimum requirement for inclusion complex formation is that the drug molecules must fit entirely or partially into the β-cyclodextrin cavity (Connors, 2000). The pKa value of mefenamic acid and the pH of solution also influence the forming of inclusion complexes. At higher pH value, mefenamic acid will more ionized. The complex of an ionic mefenamic acid

is much less stable than that of a non ionized one so it can influence solubility of mefenamic acid (International Specialty Products, 2002).

Fig. 1. Structure of mefenamic acid

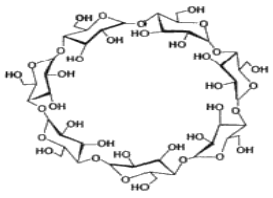
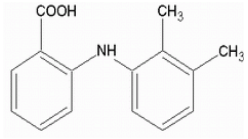


Fig. 2. Structure of β-cyclodextrin



MATERIALS AND METHODS

Materials

Mefenamic acid was purchased from Shangyu Forever Chemical Ltd. β-cyclodextrin was purchased from Sigma Aldrich. Other chemical reagents, solvent and 0.45µm membrane filter were purchased from commercial suppliers. Phosphate buffer (pH 7.0 and 8.0) and borate buffer (pH 9.0 and 10.0) were prepared in laboratory.

Method

Determination of λ_{max}

Calibration curve was made by using working solution of mefenamic acid which concentrations are 5.0mg/L-15mg/L in solutions of pH 7.0, pH 8.0, pH 9.0, pH 10.0 (with or without β-cyclodextrin $2x10^{-3}$M). Determination of λ_{max} mefenamic acid by Spectrophotometer UV-Visible Cary 50 Concentration was using two concentrations of working standard for each solution.

Solubility of mefenamic acid in aqueous solution

Excess mefenamic acid was added to aqueous solution. Solution then was sonicated with frequency of 35 kHz for 60 minutes. After sonication, the solution was shaked with velocity 110 times/minutes in water bath at temperature of 37 ± 0.5^0C for 7 hours. 5.0 mL of the solution was drawn at the 2, 4,5, 6 and 7-h. Solution was filtered using 0.45µm membrane filter then analyzed using Spectrophotometer UV-Visible at λ_{max}.

Solubility of mefenamic acid in β-cyclodextrin $2x10^{-3}$M

Excess mefenamic acid was added to the solution pH 7.0, 8.0, 9.0 and 10.0 (with or without β-cyclodextrin $2x10^{-3}$M). The solutions then were sonicated with frequency 35 kHz for 60 minutes. After sonication, the solutions were shaked with velocity 110 times/minutes in water bath at temperature 37 ± 0.5^0C until equilibration time. Solution then filtered using 0.45µm membrane filter and analyzed using Spectrophotometer UV-Visible at λ_{max}.

RESULTS AND DISCUSSION

Determination of λ_{max}

The result of the determination of λ_{max} has no difference with or without β-cyclodextrin $2x10^{-3}$M added in pH 7.0, 8.0, 9.0, 10.0. Determination of λ_{max} in 0.1N NaOH was used for phase solubility studies.

Table 1.The determination of λ_{max} mefenamic acid at various pH

Buffer Solution	λ_{max} (nm)	
	Without β-cyclodextrin $2x10^{-3}$M	With β-cyclodextrin $2x10^{-3}$M
pH 7.0	284.07	284.07
pH 8.0	284.94	284.94
pH 9.0	284.07	284.07
pH 10.0	285.04	285.04

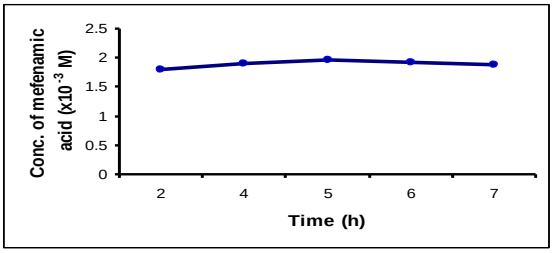
Solubility of mefenamic acid in aqueous solution

From Anova and LSD test of solubility of mefenamic acid in aqueous solution, it concluded that equilibration time was obtained at point 5-hours.

Table 2. Solubility studies in aqueous solution

Time (h)	Conc. of mefenamic acid (M)±SD
2	$1.79x10^{-3} \pm 0.0000$
4	$1.91x10^{-3} \pm 0.0000$
5	$1.96x10^{-3} \pm 0.0000$
6	$1.92x10^{-3} \pm 0.0000$
7	$1.88x10^{-3} \pm 0.0000$

Fig. 3. Equilibration time of mefenamic acid in



aqueous solution

Solubility of mefenamic acid in β-cyclodextrin $2x10^{-3}$M

The solubility of mefenamic acid at various pH were shown in Table. 3

Table 3. The solubility of mefenamic acid at various pH

Buffer solution	Conc. of mef. acid (M) ±SD		% Change of Solubility
	Without β-CD $2x10^{-3}$M	With β-CD $2x10^{-3}$M	
pH 7.0	$7.35x10^{-4}$ $\pm1.11x10^{-5}$	$9.34x10^{-4}$ $\pm1.56x10^{-5}$	27.07
pH 8.0	$1.43x10^{-3}$ $\pm4.71x10^{-6}$	$1.81x10^{-3}$ $\pm2.16x10^{-5}$	26.57
pH 9.0	$2.18x10^{-2}$ $\pm9.43x10^{-5}$	$2.25x10^{-2}$ $\pm3.30x10^{-4}$	3.20
pH 10.0	$5.51x10^{-2}$ $\pm1.89x10^{-5}$	$5.32x10^{-2}$ $\pm4.92x10^{-4}$	-3.45

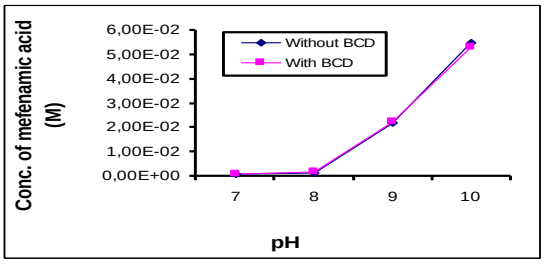


Fig. 4. Percentage solubility of mefenamic acid in base solution

Conclusion

From the results, it showed that at higher pH value, there were no significant changes by adding β-cyclodextrin $2x10^{-3}$M because there were much ionized mefenamic acid. It concluded that pKa value affected the performing of inclusion complexes of mefenamic acid-β-cyclodextrin in base solution.

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