

# Phase Solubility Studies Of Metformin Inclusion Complexes With Various Polymers Of B-Cyclodextrin And Its Derivatives

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## Abstract

With the help of inclusion complexes formed with  $\beta$ -cyclodextrin ( $\beta$ -CD), hydroxypropyl  $\beta$ -cyclodextrin (HP- $\beta$ -CD), and randomly methylated  $\beta$ -cyclodextrin (RAMEB), respectively, the current study aims to assess the potential for improving the solubility of metformin (MET), an oral antidiabetic drug, in water. It also estimates their composition and apparent stability constants,  $K_{st}$ , from phase-solubility diagrams. We have observed that the phase solubility diagrams of MET -  $\beta$ -CD and RAMEB are B type, whereas the ones for the MET - HP- $\beta$ -CD inclusion complex are A type. Since the zwitterionic structure of MET is lipophilic, all calculations were carried out in buffered phosphate solutions (pH 6). The phase-solubility diagrams show that the presence of  $\beta$ -CD and its derivatives, HP- $\beta$ -CD and RAMEB, enhances the solubility of MET to varying degrees depending on the type of cyclodextrin.

**Keywords:** Drug development, drug carriers, drug design, polymers,  $\beta$ -cyclodextrin, inclusion complexes, phase solubility diagrams, *in vitro* study.

## INTRODUCTION

The cyclodextrins capacity to prepare a hydrophobic cavity in an aqueous solution for a hydrophobic guest molecule or hydrophobic moieties in the guest molecule is what allows them to form inclusion complexes with a range of chemical molecules [1]. These inclusion complexes are important because they can increase the stability of the guest molecule or improve the aqueous solubility of the drug substances. They can also be used to control drug release, which has many potential applications in drug formulations. These inclusion complexes are widely used in the pharmaceutical domain [2]. The improvement of a drug's water solubility is particularly crucial to its bioavailability [3]. For a very long time, type 2 diabetes has been widely treated with metformin (MET) and its variants. It has no pharmacological or molecular similarities to any other class of oral antihyperglycemic drugs, and it decreases plasma glucose both basally and postprandial (that is, after eating a meal). MET is a crystalline powder that is white, hygroscopic, and bitter in flavour. Its mechanism of action and applications are comparable to those of other biguanides. Chemically, it is 1,1 dimethyl-biguanide hydrochloride [4]. This little molecule is nearly insoluble in ether or chloroform, although it is soluble in water and 95% alcohol. For a number of years, its structure was typically shown in an incorrect tautomeric form; however, this was fixed in 2005. It is low water solubility (2/20 mL of water at 37°C) and low lipophilicity ( $\log P = -0.97$ ) [5]. The chemical structure of MET is presented in Fig 1.

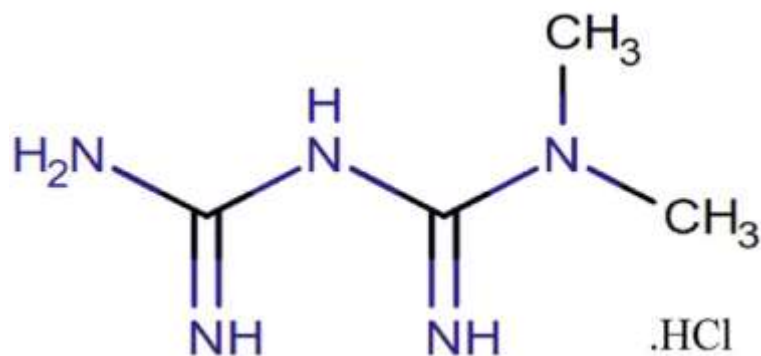


Fig 1. Chemical structure of MET hydrochloride

To assess their impact on MET solubility in water, we have examined MET's capacity to form inclusion complexes with  $\beta$ -cyclodextrin ( $\beta$ -CD), hydroxypropyl  $\beta$ -cyclodextrin (HP  $\beta$ -CD), and randomly methylated  $\beta$ -cyclodextrin (RAMEB). These inclusion complexes' synthesis and characterisation are described in another place [6]. The molar ratio and apparent stability constants of the inclusion complexes generated could be estimated with the use of the phase-solubility diagrams produced using the Higuchi and Connors approach [7].

## MATERIALS AND METHODS

### Materials

We acquired 97.69% pure metformin (MET) from SRL Pvt. Ltd, Maharashtra, India. Beta-cyclodextrin was acquired from BASF Corporation, Mumbai, India; Fluka (Sigma - Aldrich, Bangalore, India) provided 2-hydroxypropyl- $\beta$ -cyclodextrin and the randomly methylated- $\beta$  cyclodextrin (substitution rate 1.7-1.8). Analytical grade sodium hydroxide and phosphoric acid were utilised to prepare the buffers. The solvent for the chromatography was deionized ultrapure water.  $\text{H}_3\text{PO}_4$  85% solution and NaOH 1 M solution were used to create 10 mM phosphate buffer solutions. Using a Metrohm 716 DMS Titrino potentiometer, the pH values (3.00, 4.00, 5.00, 6.00, 7.00, and 8.00) were altered under potentiometrical control. We employed a Agilent Cary 60 UV-Vis Spectrophotometer, USA and a Vortex-Genie®2 (Scientific Industries) shaker for the solubility tests. Three duplicates of each experiment were carried out.

### Phase solubility study

Aqueous solutions of cyclodextrins with increasing concentrations ( $10^{-3}$  M to  $10^{-1}$  M for HP- $\beta$ -CD and  $2.5 \cdot 10^{-3}$  M to  $2 \cdot 10^{-2}$  M for  $\beta$ -CD and RAMEB) were filled with excess drug (about 100 mg MET) and shaken at  $25 \pm 0.5^\circ\text{C}$ . The best time to mix was determined by shaking the vials for 24 or 48 h, respectively. There were no discernible differences in the comparison results. The shaking time was therefore adjusted to 24 h. Following a 24-h mixing period, the specimens underwent filtration over a  $0.2 \mu\text{m}$  Nylon filter membrane (Whatman® Puradisc™), and the concentration of the dissolved MET was ascertained by measuring the absorbance at  $\lambda$  232 nm [8].

## RESULTS AND DISCUSSION

With two protonation sites and four potential species in equilibrium with one another, MET is an acid-base ampholyte. Potentiometric titration yielded the following pKa values:  $\text{pK}_{\text{a}1} = 5.22 \pm 0.07$  and  $\text{pK}_{\text{a}2} = 7.13 \pm 0.06$ . MET exists in two neutral forms (zwitterionic and uncharged form) at the isoelectric pH 5.50, which contributes to its considerable lipophilicity [9]. We have looked into how pH affects MET solubility both with and without cyclodextrins in order to determine the ideal pH of the solution utilised for the solubility studies. Using the previously obtained specific absorbance ( $A^{1\%}_{1\text{cm}} = 62$ ), the MET concentration was computed. As shown in Figs. 2a and 3b, cyclodextrins do not obstruct the measurement.

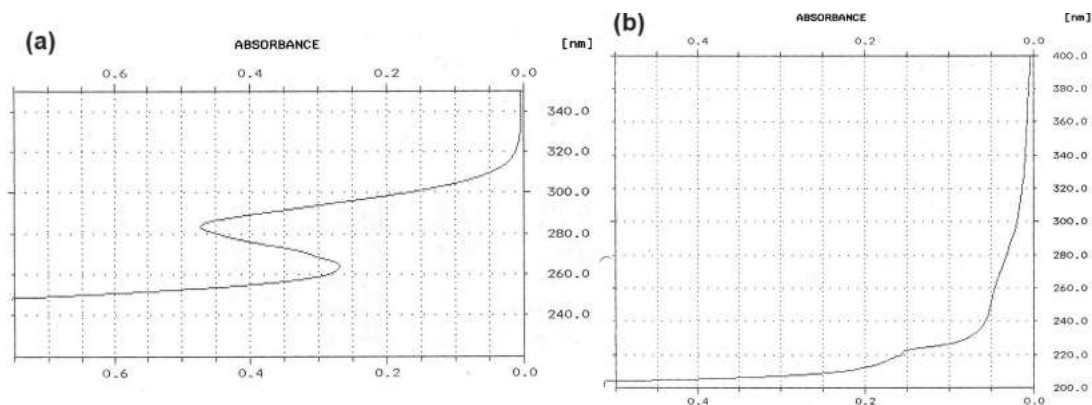


Fig 2. UV-Spectrum of (a) MET HCl at 0.5 mg/mL and (b) HP-  $\beta$ -CD at 0.1 M

As illustrated in Fig. 3, the impact of CDs on MET solubility is noteworthy throughout the pH range of 5–7; however, above pH 5.7, a combined effect of both CDs and pH takes place. It can be shown that pH ranges between 4 and 6 are where MET is least soluble. As the complexation is greatly influenced by the hydrophobicity of the MET, our studies were conducted using the methodology described by Higuchi and Connors [10] in buffered phosphate solutions at a pH of 5.7, which is also the pH range where the CDs effect on solubility is predominant (also, the pH of the mutual mixture of MET and  $\beta$ -cyclodextrin and its derivatives is 6.1–6.7).

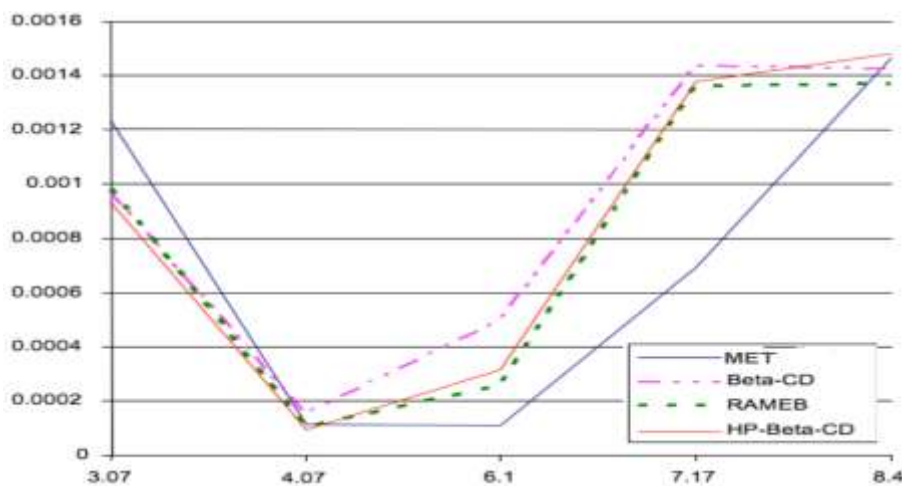
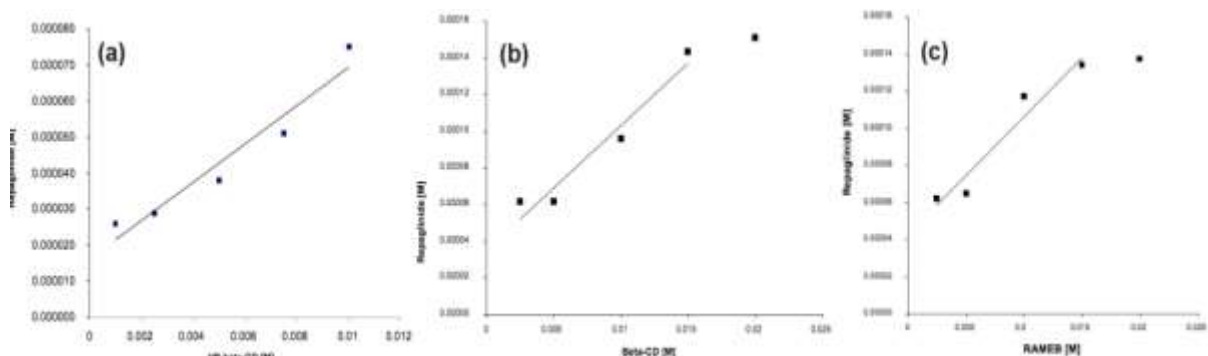


Fig. 3. The impact of pH and CDs on the solubility of MET in water

### Phase-solubility diagrams

The phase solubility diagram for the MET – HP- $\beta$ -CD inclusion complex is A type, indicating the creation of a soluble complex, in accordance with the Higuchi and Connors classification [6]. Plot shape suggests that, at very low doses of HP- $\beta$ -CD, a distinct mechanism of solubilization is likely at play (Fig 4a). When CD concentrations are high enough—above 0.01 M, roughly—complexation can happen [11]. We were able to infer that the majority of the aqueous solution contains a 1:1 inclusion complex based on the calculated regression parameters (slope =  $10^{-6}$ ). Regarding the inclusion complexes involving MET and either  $\beta$  CD or RAMEB, the phase solubility diagrams are B type. This suggests that MET's solubility increases as cyclodextrin concentration rises to approximately  $1.5 \cdot 10^{-2}$  M, after which it reaches a plateau that is likely associated with the two complexes' limited solubility (Fig. 4b and 4c).



**Fig. 4.** Phase solubility diagram of HP-  $\beta$ -CD ( $K_{st} = 354.7 \text{ M}^{-1}$ ),  $\beta$ -CD complex ( $K_{st} = 151.4 \text{ M}^{-1}$ ), and RAMEB complex ( $K_{st} = 189.2 \text{ M}^{-1}$ ) complex

The solubility curve's shape suggests that the formation of inclusion complexes with a 1:1 molar ratio is most likely (slope =  $7.1 \cdot 10^{-3}$  for  $\beta$ -CD and  $6.6 \cdot 10^{-3}$  for RAMEB) [9]. Assuming that 1:1 complex was initially generated, we estimated the apparent stability constants,  $K_{st}$ , using the parameters of the linear portion of the solubility diagrams [12].

Based on the Higuchi-Connors equation:

$$\text{Based on the Higuchi – Connors equation: } \frac{\text{slope}}{(1 - \text{slope})}$$

The stability constants for the MET – HP- $\beta$ -CD complex, MET –  $\beta$ -CD complex, and MET – RAMEB complex are determined to be  $K_{st} = 354.7 \text{ M}^{-1}$ ,  $151.4 \text{ M}^{-1}$ , and  $189.2 \text{ M}^{-1}$ , respectively. Fig. 4c presents the most likely molecular model for the 1:1 complex MET: HP- $\beta$ -CD.

## CONCLUSIONS

$\beta$ -CD, along with its derivatives HP- $\beta$ -CD and RAMEB, improves the solubility of MET, a medication that is nearly water insoluble. With MET, the CDs under study create a 1:1 inclusion complex. When  $\beta$ -CD and RAMEB are present, inclusion complexes with restricted solubility are generated, whereas HP- $\beta$ -CD yields a soluble complex. Based on the determined apparent stability constants, the MET complexes with  $\beta$ -CD, HP- $\beta$ -CD, and RAMEB exhibit the following variations in stability: RAMEB >  $\beta$ -CD > HP- $\beta$ -CD.

## Declarations

## Acknowledgments

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## Ethical Approval

Not Applicable in this section.

## Competing Interest

Author declares that there is no potential conflict of interest in this paper

## Author Contribution

All the authors are equally contributed and approved the manuscript.

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