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A study on the supramolecular structure of inclusion complex of β-cyclodextrin with prazosin hydrochloride

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Abstract

The supramolecular structure of the inclusion complex of β-cyclodextrin (βCD) with prazosin hydrochloride (PRH) has been investigated. The 1:1 stoichiometry of complexation was achieved by phase solubility study and the inclusion complex with 1:1 molar ratio was prepared by co-precipitation method. DSC analysis confirmed that the inclusion complex was formed. XRD patterns of βCD and inclusion complex indicated that the structures of them belonged to the channel-type packing structure, and the axial structure period was two times the depth of βCD torus. The fragment of PRH molecule which entrapped into the βCD cavity was proposed on the basis of Fourier transformation-infrared analysis and the predicted model indicated that the furan ring and the conjunctive carbonyl group of PRH molecule were inserted into the βCD cavity.

Keywords: β-Cyclodextrin; Prazosin hydrochloride; Inclusion complex; Supramolecular structure; Channel-type packing structure

1. Introduction

Cyclodextrins (CDs) are a family of the macrocyclic oligosaccharides known as α-cyclodextrin, β-cyclodextrin (βCD), and γ-cyclodextrin, which are composed of six, seven, or eight α- (1,4) linked glycosyl units, respectively. βCD (Fig. 1a) is the most accessible, the lowest-priced and generally the most useful. The cyclodextrin structure provides a molecule shaped like a segment of a hollow cone with an exterior hydrophilic surface and interior electron-rich hydrophobic cavity of 0.79 nm depth (Rekharsky & Inoue, 1998), which is capable of forming stable, supramolecular structures with various molecules (Liu & Zhu, 2006; Sanghavi, Mayekar, & Fruitwala, 1995; Torres-Labanderia, Blanco-Mendez, & Villa-Jato, 1994), fitting partially or completely in the host molecular cavity.

Prazosin hydrochloride (PRH, Fig. 1b), 1-(4-amino-6, 7-dimethoxy-2-quinazolinyl)-4-(2-furanylcarbonyl) piperazine HCl, which was a selective α_1-adrenoceptor antagonist in clinical use, has gained a widespread acceptance in the management of hypertension and in the treatment of congestive heart failure (Solomon, Wier, Ippolito, & Toscano, 1997; Sreedhar, Sastry, Reddy, & Sankar, 1996).

In this paper, the inclusion complex of βCD with PRH was obtained by co-precipitation method, a widely used method for the preparation of inclusion complex. Phase solubility study of the complex in solution was conducted to determine the stoichiometry of complexation. The supramolecular structure of the 1:1 inclusion complex between βCD and PRH was achieved by differential scanning calorimetry (DSC), X-ray diffractometer (XRD) and Fourier transformation-infrared spectroscopy (FTIR) for understanding which fragment of PRH molecule was involved into the βCD cavity.

2. Experimental section

2.1. Materials

PRH ($M_r = 420$) was obtained from Songsheng Institute and Plant of Pharmaceutical and Chemical (Jiangsu,

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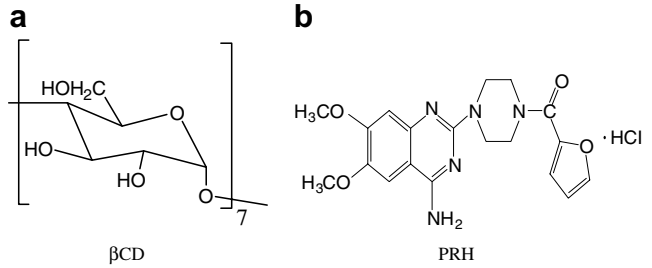


Fig. 1. Chemical structure of βCD and PRH.

China), βCD ($M_r = 1135$) from Shanghai Bio Life Science and Technology Co. Ltd (Shanghai, China). All the reagents were of analytical grade. Distilled water was used all through the experiment.

2.2. Phase solubility study

Phase solubility study was carried out in distilled water according to the method discussed by Higuchi and Connors (1965). In brief, excess amount of PRH (40 mg) was added to 10 ml of aqueous solutions containing various concentrations of βCD (0–0.016 M). Then the suspensions were sealed and shaken at (25 ± 2) °C for 7 days. After equilibrium attainment, the samples were filtered through 0.45 μm membrane filter and properly diluted. The concentration of PRH was determined spectrophotometrically (S52, China) at 246 nm (USP 26). The presence of βCD did not interfere with the spectrophotometry assay of PRH. Each experiment was performed in triplicate. The apparent stability constant K_s was calculated from the phase solubility diagram, with the assumption of 1:1 stoichiometry, according to the following equation:

$$K_s = \frac{\text{slope}}{S_0(1 - \text{slope})} \quad (1)$$

S_0 is the solubility of PRH in the absence of βCD.

2.3. Preparation of binary systems

The inclusion complex of PRH with βCD at 1:1 molar ratio was prepared using the co-precipitation method. The accurately weighed βCD was dissolved in distilled water to get a saturated solution. Then, PRH solution in methanol was added slowly, a suspension was formed. The suspension was stirred at 40 °C for 30 min and maintained stirring at room temperature for 24 h. The obtained mass was filtered through 0.45 μm membrane filter and dried at 40 °C in an oven for 24 h. The dried complex was ground to fine powder and screened through an 80-mesh sieve.

Physical mixture of PRH and βCD with 1:1 molar ratio was prepared by mixing exactly weighed amount of PRH and βCD for 20 min in a mortar. The mixture was passed through an 80-mesh sieve before use.

2.4. Characterization

DSC was carried out in the temperature range of (50–300) °C in a stream of nitrogen atmosphere on a Perkin-Elmer DSC7 differential scanning calorimeter with a Pyris Series workstation (Perkin-Elmer, USA). The accurately weighed sample was placed in an aluminum pan, and an empty aluminum pan was used as reference. The scanning rate was 10 °C/min and the nitrogen flow was 20 ml/min.

The powder X-ray diffraction was recorded using XD-98 diffractometer, operated at a voltage of 40 kV and a current of 36 mA. The samples were analyzed in the 2θ angle range of (3–40)° and the process parameters were set as: scan step size of 0.02°, scan step time of 1.54 s.

Infrared spectrum of the inclusion complex was obtained using a Shimadzu FTIR-8900 spectrometer (Shimadzu, Japan) according to potassium bromide disk method. The IR spectra of pure PRH, βCD as well as their physical mixture of 1:1 molar ratio were also obtained by the same procedure for comparison. The scans were executed at a resolution of 8 cm^{-1}, from 4000 to 400 cm^{-1}.

3. Results and discussion

3.1. Phase solubility study

This study allowed us to follow the inclusion phenomena and evaluate the apparent stability of the complex. The phase solubility profile of PRH–βCD was presented in Fig. 2, which could be classified as Bs-type according to Higuchi and Connors. The solubility of PRH increased with the increment of the βCD concentration in the range of (0–8) mM. When the concentration of βCD was more than 8 mM, it would lead to the formation of a water-insoluble substance (Manolikar & Sawant, 2003). The 1:1 molar ratio of the inclusion complex was achieved from the initial

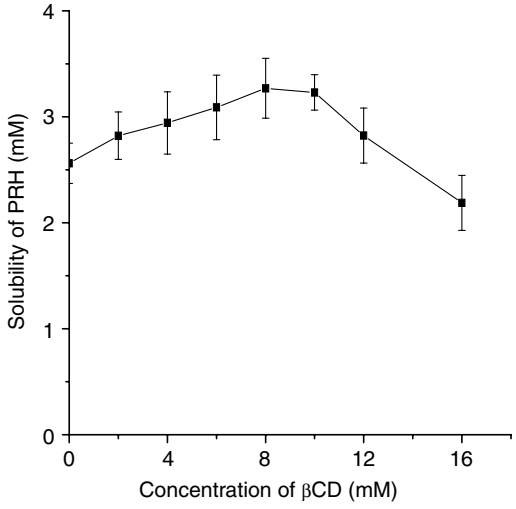


Fig. 2. Phase solubility diagram of PRH–βCD system in water.

ascending part of the curve, a nearly straight line with the slope of 0.084 ($r = 0.9919$). The apparent stability constant $K_{1:1}$ was obtained to be $29.7 \pm 2.5\ M^{-1}$ according to Eq. (1). Such result was in agreement with those obtained from other inclusion complexes where the βCD was bound to the small hydrophobic molecules (Fernandes, Vieira, & Veiga, 2002).

3.2. DSC

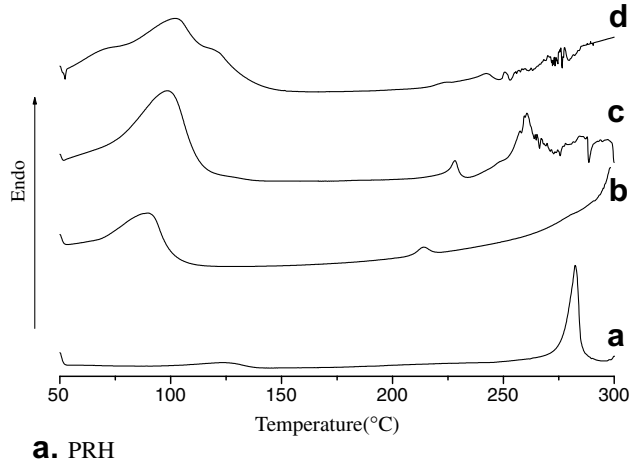
DSC can be used for the recognition of inclusion complexes. When guest molecules were embedded into βCD cavities, their melting, boiling or sublimating points generally shifted to different temperatures or disappeared (Marques, Hadgraft, & Kellaway, 1990). The thermograms of βCD, PRH and their binary systems were shown in Fig. 3.

The DSC diagram of PRH (Fig. 3a) exhibited a sharp endothermic peak at 282.3 °C, indicating the melting point of PRH. The trace of βCD (Fig. 3b) showed a very broad endothermic effect between 60 and 110 °C, which attained a maximum around 90 °C, corresponding to the dehydration process, followed by an irreversible solid–solid phase transition at 214 °C and finally to a degradation process, which took place at around 300 °C (Giordano, Novak, & Moyano, 2001). For the physical mixture of PRH and βCD (Fig. 3c), the drug endothermic peak shifted from 282.3 to 260.6 °C and the peak of βCD from 214 to 228.2 °C. While turning to the inclusion complex prepared by co-precipitation method (Fig. 3d), there was only a dehydrated peak of βCD, the endothermic peak of PRH at 282.3 °C disappeared, suggesting that a certain fraction of PRH molecule was included into the βCD cavity.

3.3. XRD

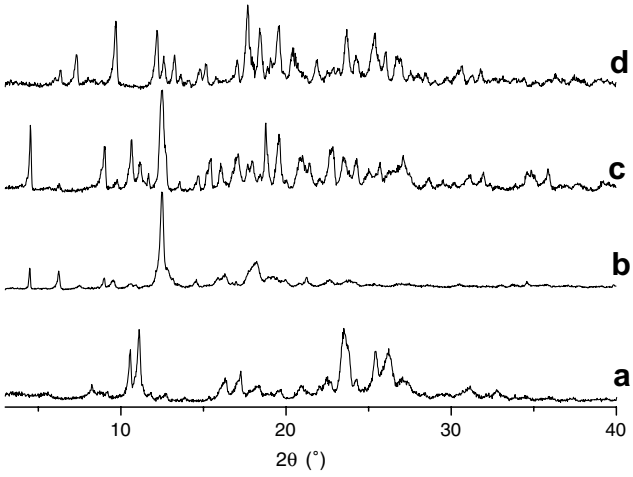
XRD was used to determine the structure period of inclusion complex. Crystals of βCD could be mostly categorized into two types, cage-type and channel-type (Yong, Li, Zheng, & Zhou, 2000). In cage-type structure, βCD molecules are arranged in a herringbone fashion, and both ends of the βCD cavity are blocked by adjacent molecules (Harata & Kawano, 2002). The channel-type structure is formed by the linear stack of βCD rings. The column-like structure can include molecules that are longer than the depth of the βCD cavity and penetrating two or more βCD rings.

Fig. 4b was the X-ray diffraction pattern of βCD. The most intense peak of βCD (relative intensity $I/I_0 = 100$) was in the position of $2\theta = 12.5°$ ($d = 0.71$ nm, the depth of the βCD cavity), while in the position of $2\theta = 6.2°$, the d value was 1.41 nm, two times the depth of the βCD cavity, which could be defined as channel-type packing and the axial structure period was two times the depth of βCD torus. The channel-type structure is further classified into two types, the head-to-head and the head-to-tail. Only head-to-head type was in accordance with the axial structure period of βCD with two times of the cavity height (Yong et al., 2000). The peaks with the same d values in position of $2\theta = 12.5°$ and $2\theta = 6.2°$ also appeared in the XRD pattern of the inclusion complex (Fig. 4d) between βCD and PRH, which could be concluded that no change was made in the cavity structure of βCD after formation of inclusion complex. However, the most intense peak in the XRD pattern of inclusion complex showed in the position of $2\theta = 17.9°$, while the most intense peak in the XRD



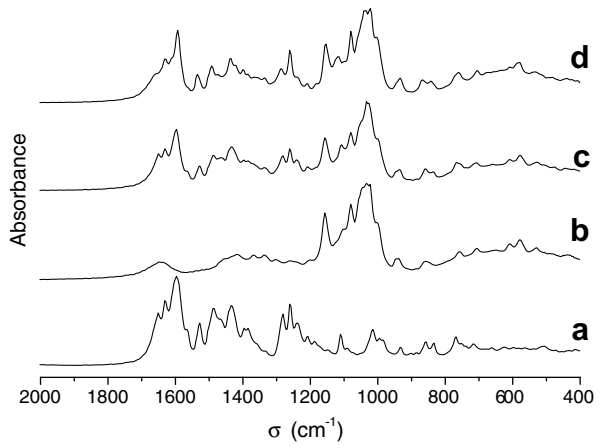
a. PRH
b. βCD
c. physical mixture
d. inclusion complex

Fig. 3. DSC diagram of PRH–βCD systems.



a. PRH
b. βCD
c. physical mixture
d. inclusion complex

Fig. 4. XRD pattern of PRH–βCD systems.



a. PRH

b. βCD

c. physical mixture

d. inclusion complex

Fig. 5. FTIR spectra of PRH–βCD systems.

pattern of PRH (Fig. 4a) was in the position of $2\theta = 11.1°$, which indicated the formation of inclusion complex between βCD and PRH.

3.4. FTIR

Fig. 5 showed the infrared spectra of wave number from 2000 to 400 cm^{-1} of βCD, PRH, the physical mixture of βCD and PRH at 1:1 molar ratio as well as the inclusion complex.

PRH showed a very strong absorption band between 1720 and 1560 cm^{-1} for carbonyl stretching band, which split into triplet (the absorption peak showed in 1650, 1631 and 1593 cm^{-1}, respectively) due to the influence of the furan ring and the atom N in the piperazine ring attached to the carbonyl group. 1261 and 1434 cm^{-1} were for ether absorption band of ph–O–C and C–H in the aromatic ring, respectively. 1527 cm^{-1} was denoted for stretching vibration of C=C in the aromatic ring. 1107 cm^{-1} was corresponding to the characteristic band of C–O–C stretching vibration in the furan ring. Inclusion complex of PRH–βCD did not show any new peaks, indicating no chemical bonds were created in the formed complexes.

The characteristic triplet appeared also at the same position in the physical mixture. However, the spectrum of inclusion complex, whose band changed to doublet with the peak in 1650 cm^{-1} disappeared, suggested that the carbonyl group of PRH was entrapped into the host cavities, during inclusion complexation. The band of C–O–C stretching vibration in the furan ring (1107 cm^{-1}), observed for the physical mixture, shifted to 1118 cm^{-1} for inclusion complex, indicating the restrict in infrared vibration after formation of inclusion complex. Other bands such as 1527 cm^{-1} suffered a slight shift, probably due to the influence of overlapping with the βCD in the same zone, which perturbs the energy of these vibrations.

These results indicated that the vibrating and bending of the PRH molecule was restricted due to the formation of inclusion complex, the furan ring and the conjunctive carbonyl group were inserted into the cavity of βCD molecule.

3.5. Molecular modeling

The structural model of PRH–βCD inclusion complex was established on the basis of the results of phase solubility, DSC, XRD and FTIR measurements. According to the phase solubility study, a 1:1 stoichiometry of PRH–βCD was found. DSC analysis confirmed that the inclusion complex was formed. The structure of inclusion complex was defined as head-to-head channel-type packing whose axial structure period of inclusion complex was two times the depth of βCD cavity according to the XRD study. It was concluded that the inclusion effect of βCD with PRH occurs between βCD cavity and the furan ring as well as the adjacent carbonyl group of PRH molecule from FTIR analysis. The longest distance between different atoms of furan ring was calculated to be 0.51 nm with the information of bond lengths and angles, and the inner diameter of the βCD cavity in the narrow edge and wide edge were 0.60 and 0.68 nm, respectively (Szejtli, 1988). According to the size matching rule (Liu, Chen, Li, Wada, & Inoue, 2001), the fragment of PRH molecule mentioned above was entrapped into the narrow edge of βCD cavity. Therefore, the predicted model of PRH–βCD inclusion complex was proposed and shown in Fig. 6.

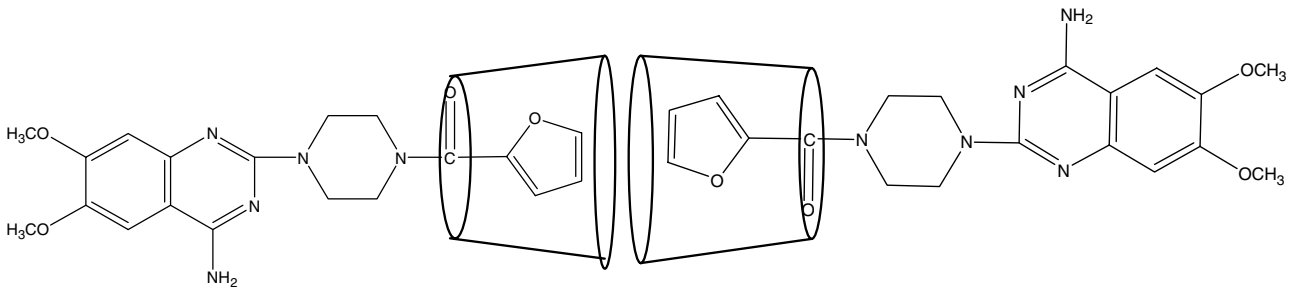


Fig. 6. Proposed structure model for PRH–βCD inclusion complex.

4. Conclusions

In summary, the inclusion complex with 1:1 molar ratio was formed between βCD and PRH. The furan ring and the conjunctive carbonyl group of PRH molecule were inserted into the βCD cavity.

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