[Chem. Pharm. Bull. 32(10)4179—4184(1984)]

Inclusion Complexes of Cinnarizine with β-Cyclodextrin in Aqueous Solution and in the Solid State[1)]

TADAKAZU TOKUMURA,*,[a] HARUHISA UEDA,[b] YUKI TSUSHIMA,[a] MASAYOSHI KASAI,[a] MASANORI KAYANO,[a] IWAO AMADA[a] and TSUNEJI NAGAI[b]

Research Laboratories of Technological Development, Eisai Co., Ltd.,[a] 2–3–14, Minami, Honjo-shi, Saitama 367, Japan and Faculty of Pharmaceutical Sciences, Hoshi University,[b,2)] Ebara 2–4–41, Shinagawa-ku, Tokyo 142, Japan

(Received February 9, 1984)

Inclusion complex formation of cinnarizine with β-cyclodextrin in aqueous solution and in the solid state was confirmed by the solubility method, powder X-ray diffractometry, differential scanning calorimetry (DSC) and proton nuclear magnetic resonance (^{1}H-NMR) spectroscopy. A solid complex of cinnarizine with β-cyclodextrin in 1 : 2 molar ratio was prepared by a coprecipitation method and a neutralization method, and its dissolution behavior in pH 5.0 buffer solution was examined in comparison with those of several other cinnarizine complexes. The dissolution rate of cinnarizine in the inclusion complex was about 30 times larger than that of intact cinnarizine, being almost the same as that of cinnarizing/PVP K-30 complex (weight ratio 1 : 5).

Further, a new and efficient procedure was developed to prepare cinnarizine/β-cyclodextrin inclusion complex on a manufacturing scale.

Keywords——cinnarizine/β-cyclodextrin inclusion complex; phase solubility method; dissolution profile; powder X-ray diffractometry; differential scanning calorimetry; ^{1}H-NMR

Cinnarizine [1-(diphenylmethyl)-4-(3-phenyl-2-propenyl)-piperazine], an agent for increasing cerebral blood flow, is widely used orally to treat various problems in cerebral apoplexy, cerebral arteriosclerosis and post traumatic cerebral symptoms. However, cinnarizine ($pK_a = 7.47$, weak base)[3)] is only very slightly water-soluble (the solubility at 37 °C is less than 10 mg/100 ml). Therefore, it is poorly absorbed in patients with achlorhydria.[4)] Cyclodextrin complexation has been extensively applied to enhance the solubility,[5)] dissolution rate,[6)] and bioavailability[7)] of slightly soluble drugs. Thus, this investigation was carried out with the aim of improving the dissolution characteristics. The formation of an inclusion complex of cinnarizine with β-cyclodextrin was confirmed by the solubility method, powder X-ray diffractometry, differential scanning calorimetry (DSC) and proton nuclear magnetic resonance (^{1}H-NMR) spectroscopy.

Experimental

Materials——Cinnarizine (CN) and β-cyclodextrin (β-CD) were obtained from Eisai Co., Ltd., and Nippon Shokuhin Kako Ltd., respectively. All other chemicals and solvents were of analytical reagent grade. Distilled water for injection was used throughout the study.

Solubility Studies——CN (30 mg, 4.07×10^{-3} M) was added to 30 ml of water or β-cyclodextrin solution (0.05 to 2.0×10^{-2} M) is a Nessler tube, which was then sealed and shaken at 20 °C. To achieve equilibrium, the sample solution was occasionally ultrasonicated (power: 150 W). After equilibration (10 d), an aliquot was filtered with a Toyo TM-2 membrane filter (0.22 μm). The concentration of CN in the filtrate was determined by the ultraviolet (UV) absorption method using a Hitachi UV-124 spectrophotometer.

Powder X-Ray Diffraction Study——Powder X-ray diffractometry was carried out using a Rigaku Denki

Geigerflex model D-2 diffractometer with Ni-filtered Cu-K_α radiation.

DSC Study——This was done using a Perkin-Elmer model 1B differential scanning calorimeter.

NMR Spectroscopic Study——This was done in 0.1 N DCl using a JNM-FX 100 NMR spectrometer; ^{1}H-chemical shifts are given relative to external tetramethylsilane within ± 0.002 ppm.

Preparation of Samples of Dissolution Study——a) CN/β-CD Complex and CN/γ-CD Complex: CN/β-CD complex was prepared by the coprecipitation method (method I) and the neutralization method (method II), as shown in Chart 1. CN/γ-CD complex was prepared by the neutralization method.

b) Cinnarizine (CN) and Polyvinylpyrrolidone K-30 (PVP K-30) Coprecipitate: Both 5 g of CN and PVP K-30 at a suitable weight ratio were dissolved in 40 ml of 1,1-dichloroethane and 100 ml of ethanol, then the solvent was removed *in vacuo* using a rotary evaporator at about 60 °C. The preparations was dried *in vacuo* at room temperature for 24 h, then ground in a mortar and stored in a desiccator.

c) CN and Crystalline Cellulose (MCC) Ground Mixture: CN and MCC in 1 : 9 weight ratio was prepared by grinding in a hammar mill (several runs).

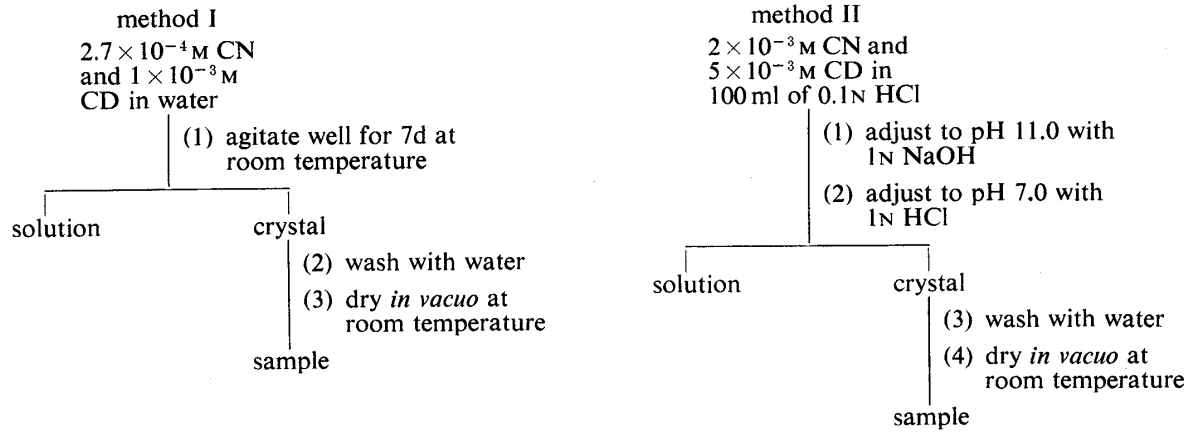


Chart 1. Methods for Mass Preparation of Inclusion Complex

Procedure for Dissolution Study——The paddle method of the Pharmacopoeia of Japan, edition X (JP X), was used in pH 5.0 buffer solution. The buffer solution was made up of the 1st fluid and 2nd fluid of JP X. A certain amount of each sample corresponding to 20 mg of CN (retained on a 100 mesh sieve and passing a 20 mesh sieve) was placed in a beaker containing the medium equilibrated to 37 °C. The paddle was rotated at 100 rpm, and the sample solution was passed through a flow cell attached to a Hitachi 156 dual-wavelength recording spectrophotometer. The concentrations of CN were determined at 300 nm (λ_1) and 254 nm (λ_2).

Results and Discussion

Phase Solubility Diagram

Complex formation of CN with β-CD was studied by a solubility method. Figure 1 shows an equilibrium phase solubility diagram obtained for the CN/β-CD system in water. The solubility of CN increased about five times in the presence of β-CD, showing the features of a B_s type phase diagram.[8] In the higher concentration range of β-CD, a solid complex was precipitated. In order to determine the CN content, the solid complex was dissolved in 50% ethanol and the concentration of CN in the solution was determined by the UV absorption method at 254 nm. The CN content of the solid complex was 14.1%. The stoichiometry of the complex was found to be 1 : 2 (CN: β-CD) from the CN content. The result is in good accordance with that based on the data in the plateau region of the solubility diagram. The apparent stability constant, K', of the complex was estimated to be $K' = 6.2 \times 10^3\ \mathrm{M}^{-1}$ from the initial straight line portion of the solubility diagram. This value is larger than the previously reported values for sulfonylurea/β-CD complexes,[9] antiinflammatory/β-CD complexes,[10] and sulfonamide/β-CD complexes.[11] This large stability constant may be a result of substantial 1 : 2 inclusion complex formation.

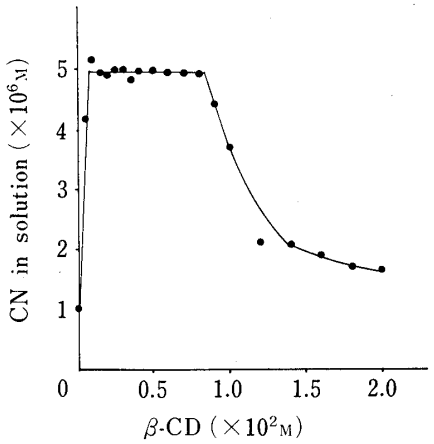


Fig. 1. Solubility of CN as a Function of β-CD Concentration in Water at 20 °C

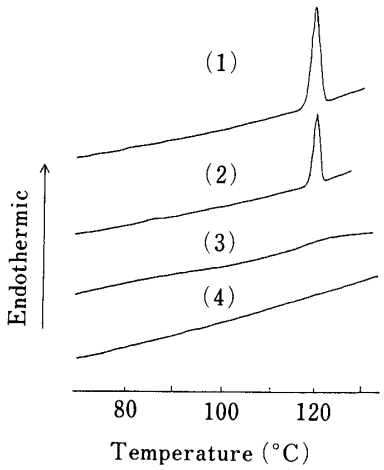


Fig. 3. DSC Curves of CN/β-CD at a Scanning Speed of 8 °C/min

1, intact CN; 2, physical mixture of CN and β-CD; 3, inclusion complex CN/β-CD (method I); 4, inclusion complex CN/β-CD (method II).

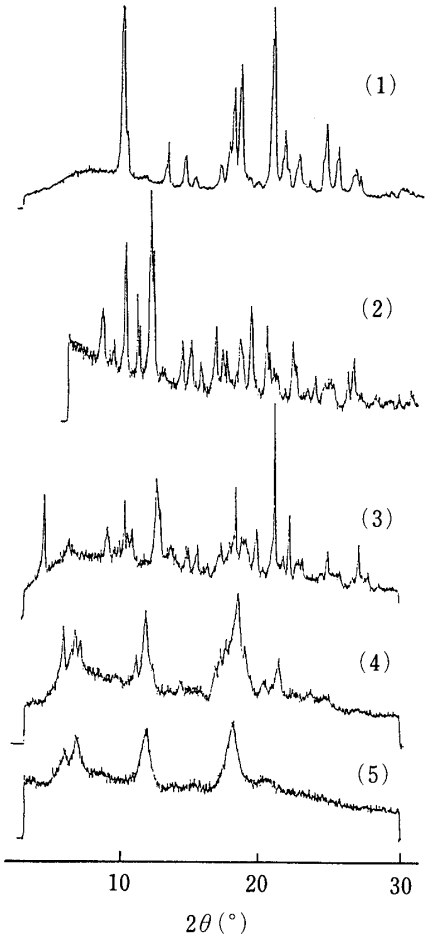


Fig. 2. Powder X-Ray Diffraction Patterns of CN/β-CD

1, intact CN; 2, recrystallized β-CD; 3, physical mixture of CN and β-CN; 4, inclusion complex CN/β-CD (method I); 5, inclusion complex CN/β-CD (method II).

Method for Preparation of Inclusion Complex on a Manufacturing Scale

The ordinary methods,[12] *e.g.* the coprecipitation method based on phase solubility (method I) and the kneading method, are not appropriate to obtain CN/β-CD complex with satisfactory efficiency and yield. However, the inclusion complex could be prepared easily by the neutralizing procedure (method II). The stoichiometry of the complex described above was then analyzed chemically, and was found to be 1 : 2 (CN: β-CD). Further, method II was experimentally adapted to the CN/α-CD system and CN/γ-CD system. The CN/γ-CD complex was obtained easily, and found to have 1 : 1 stoichiometry from the CN content (24.2%). However, no CN/α-CD complex was obtained; the α-CD cavity might be too small for complexation of CN. The principal advantages of method II can be summarized as follows: (1) the unnecessity of organic solvent, (2) a good yield in a short operating time and (3) suitability for extension to manufacturing scale.

Further Evidence of Inclusion Complexation

Figure 2 shows the powder X-ray diffraction pattern of the complexes prepared by different methods in comparison with that of a physical mixture at the same molar ratio. The

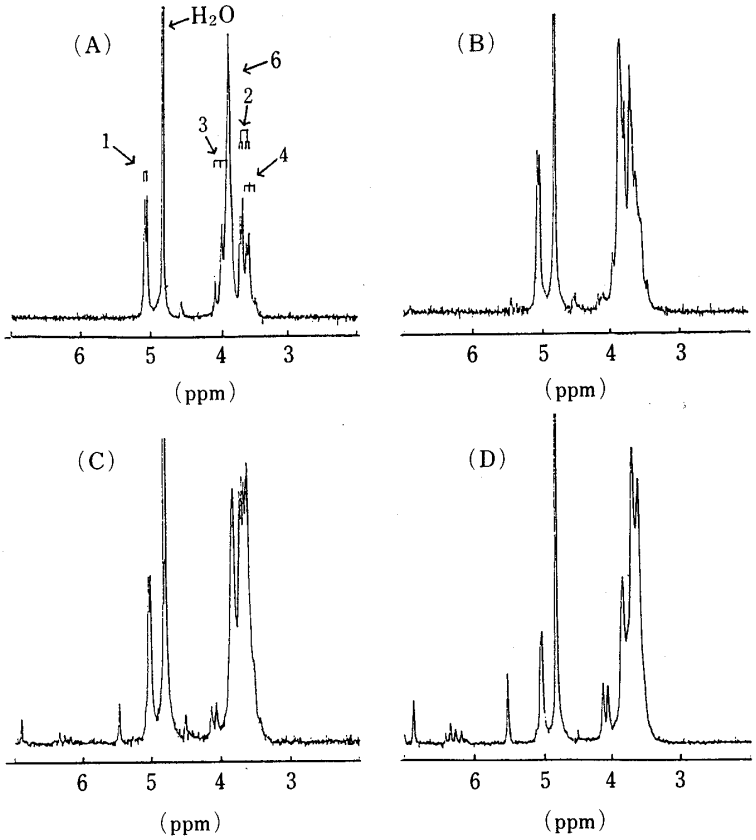


Fig. 4. ^{1}H-NMR Spectra of β-CD Containing Various Amounts of CN

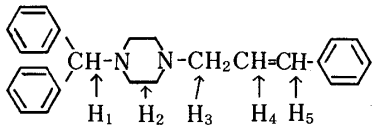
Molar ratio of CN/β-CD: (A) 0.00, (B) 0.50, (C) 1.00, (D) 2.00. Solvent: 0.1 M DCl in D_2O.

diffraction pattern of the physical mixture was found to be simple superposition of those of the components, while that of the complex was apparently different, corresponding to a new solid phase. In addition, there was no difference in diffraction patterns among the complexes. These results indicate that CN interacted with β-CD to form an inclusion complex.

Figure 3 shows the DSC curves of CN/β-CD. The endothermic peak at around 120 °C, which was observed for intact CN and the physical mixture of CN with β-CD, disappeared in the CN/β-CD inclusion complex. This result corresponded to the change in X-ray diffraction patterns.

Figure 4 shows the effect of CN on the ^{1}H-NMR spectrum of β-CD in D_2O. Protons located in the cavity of β-CD such as H-3 showed a high-field shift and progressive broadening with increasing amount of CN added. On the other hand, no anisotropic shielding was detected of protons located outside the cavity, such as H-1 and H-2.

Figure 5 shows the effect of β-CD on the ^{1}H-NMR spectrum of CN in D_2O. The chemical shifts of the phenyl signals at δ 7.6—7.3 could not be determined exactly because of the multiplet structure. Further, the signals of the H_2-protons (piperazine ring) overlapped with signals due to β-CD. It was apparent that the H_1-proton signal was remarkably shifted to higher field in the presence of β-CD, suggesting that the ring-currents of the diphenyl group in the drug were affected by the binding to β-CD. The lowfield shift of the H_3-proton might be induced by decreased freedom of rotation of the 3-phenyl-2-propenyl group due to the interaction with β-CD. This hypothesis could not be substantiated by measuring the carbon-13 nuclear magnetic resonance (^{13}C-NMR) spectrum, because the solubility of CN in water is very low. However, the phenyl groups on both sides of the CN molecule might interact with β-CD at a molar ratio of 1 : 2 (CN: β-CD). This speculation was considered to be consistent with



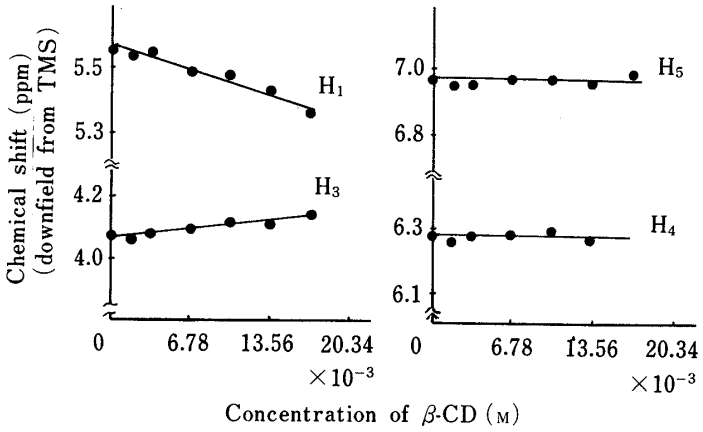


Fig. 5. Variation of ^1H Chemical Shifts of 6.78×10^{-3} M CN with Concentration of β-CD

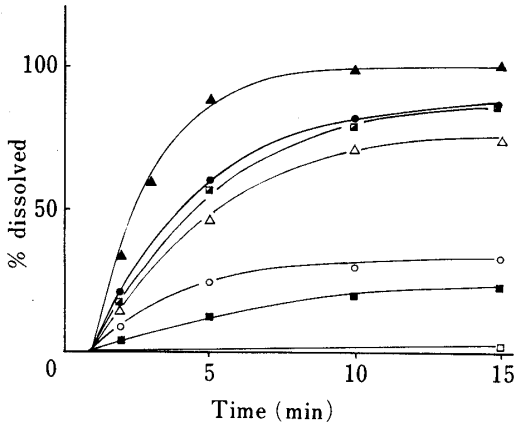


Fig. 6. Dissolution Rate Curves of CN Complexes in pH 5.0 Buffer Solution

▲, CN/PVP K-30 complex (1 : 10 weight ratio); △, CN/PVP K-30 complex (1 : 5 weight ratio); ◪, CN/β-CD complex (method I, 1 : 2 molar ratio); ●, CN/β-CD complex (method II, 1 : 2 molar ratio); ○, CN/γ-CD complex (method II, 1 : 1 molar ratio); ■, CN/MCC ground mixture (1 : 9 weight ratio); □, intact CN.

Each point is the mean of three determinations.

the results obtained by the solubility method.

Dissolution Behavior of Cinnarizine and Its Complexes

The relative rates of dissolution of CN and some complexes of CN in powder form are shown in Fig. 6. It is evident that each complexes form of CN dissolved much more rapidly than intact CN. The dissolution rates of CN/β-CD complex prepared by two different methods, method I and method II, were essentially the same, and the dissolution rate of CN/β-CD complex was 30 times larger than that of intact CN. The enhancement was similar to that obtained with CN/PVP K-30 complex (weight ratio 1 : 5). The enhancement of the dissolution characteristics of CN by β-CD inclusion complexation may be due to improvements in solubility and wettability. The above results suggest that complex formation with β-CD may enhance the bioavailability of CN.

References and Notes

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