



Relationship between Separation Behavior and Structural Flexibility in Inclusion Crystal of Cholic Acid

KAZUNORI NAKANO^{1,*}, SHIGENDO AKITA¹, NUNGRUETHAI YOSWATHANANONT², KAZUKI SADA^{2,**} and MIKIJI MIYATA²

¹Nagoya Municipal Industrial Research Institute, 3-4-41 Rokuban, Atsuta-ku, Nagoya 456-0058, Japan; ²Department of Material and Life Science, Graduate School of Engineering, Osaka University and Handai FRC, 2-1 Yamadaoka, Suita, Osaka 565-0871, Japan

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Abstract

Selective incorporation of two aromatic compounds, benzene and ethylbenzene, into an inclusion crystal of cholic acid was investigated. Addition of an excess amount of 1:1 mixture of benzene and ethylbenzene into saturated solution of cholic acid in 1-butanol led to a spontaneous formation of an inclusion crystal. The co-crystal contained benzene and ethylbenzene at the constant molar ratio of 8:2 irrespective of the relative concentrations of guest and host in the feed solution, indicating that the resulting crystal consists of the two guests mixed in a single host framework. The resulting ternary crystal had thermal behavior similar to a binary crystal obtained from benzene. In contrast to the guest/host ratio, the benzene/ethylbenzene ratio in the feed solution affected that in the inclusion crystal. Benzene was basically preferred in the cholic acid crystal over ethylbenzene, but the selectivity reversed at an excess amount of ethylbenzene. This separation behavior can be understood in terms of the structural flexibility of host frameworks.

Introduction

Considerable attention has been paid to lattice inclusion compounds as separation media for organic compounds [1]. To take an example, urea is known to form adducts with appropriate solutes, and they have been employed in separations of *n*-paraffins from branched ones. Recently, Hassan and coworkers studied adsorption of *n*-paraffins on solid urea rather than into urea crystal to improve the separation efficiency [2, 3]. Moreover, Toda and coworkers have expanded the potential of lattice inclusion crystals for separation using various host compounds such as brucine, sparteine, bis- β -naphthol, tartarate derivatives, acetylenic alcohols, and alkylammonium halide [4, 5]. Aoyama and coworkers reported the separation of ketones using anthracene-resorcinol derivatives [6]. One of the characteristic properties of these lattice inclusion compounds is flexibility of their host frameworks and cavities. Since the host frameworks are constructed by non-covalent bonds, such as hydrogen bonding, π - π stacking, and van der Waals interaction, the cavities easily change their shape and size dependent on incorporated guest compounds [7]. Therefore, lattice inclu-

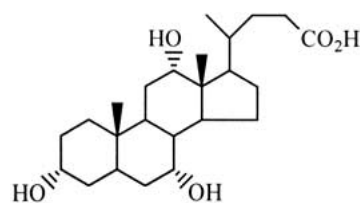


Figure 1. Molecular structure of cholic acid (CA).

sion compounds have potential for molecular recognition in a wide variety of organic compounds with high specificity. Furthermore, the mechanism of molecular recognition is distinct from that of macrocyclic host compounds with rigid cavity constructed by covalent bonds [8].

Cholic acid (CA, Figure 1), a naturally occurring bile acid, is known to form inclusion crystals consisting of host lattice frameworks with various guest compounds. In previous papers, we revealed that 1-butanol is useful as a solvent for the formation of CA inclusion compounds with various organic compounds [9]. The host frameworks of the compounds are dependent on the guest molecules and are categorized into several types [10]. The dependence of the framework types can be understood in terms of the fit of the guest molecule in the host cavity [7]. We have already reported molecular recognition of aromatic hydrocarbons from their 1:1 binary mixtures, and the mechanism of the selectivities has been demonstrated [11, 12]. In the present

* Author for correspondence.

E-mail: nakano.kazunori@nmiri.city.nagoya.jp

** Present Address: Department of Chemistry and Biochemistry, Graduate School of Engineering, Kyushu University, 6-10-1 Hakozaki, Higashi-ku, Fukuoka 812-8581, Japan.

report, we further investigated the effects of the ratios of the binary guest mixtures on the guest recognitions through the determination of the host framework type of the resulting inclusion crystals. These experiments would explain the molecular recognitions of the flexible CA host frameworks.

Experimental

Reagents

CA was purchased from Wako Co., and used without further purification. All other chemicals and solvents were of the commercially available purest grades.

Procedure

A host solution was prepared by dissolving CA (130 mg) in 1-butanol (0.4 ml), and guest mixtures were prepared by mixing two guest compounds at a predetermined molar ratio. After mixing the two solutions in a 13 ml vial, the mixture was allowed to settle overnight at 20 °C to yield crystals. Inclusion crystals thus obtained were filtered out and settled on a filter paper for some time to remove the adhering solvent and guests on the surface.

Amounts of the guests incorporated within the crystals were determined by gas chromatography (GC) using a Shimadzu GC-14A instrument after dissolving the crystal in methanol. Thermal gravimetry (TG) was performed on a SEIKO TG/DTA 200 system; *ca.* 10 mg of the inclusion crystals was heated from 40 to 230 °C at a rate of 5 °C min⁻¹. The host framework types of CA inclusion crystals were determined by X-ray powder diffraction (Rigaku RINT-1100) according to our previous systematic investigation by X-ray crystallography [7].

Results and discussion

Formation of a CA inclusion crystal from a 1:1 guest mixture

Mixing solutions of the host and guest causes spontaneous crystallization of CA, because the guest compounds decrease solubility of CA in 1-butanol. Any seeding crystals are not required. All the resulting crystals include the aromatic guest compounds, while the solvent, 1-butanol, is not included at all [9]. CA crystals without guests are not formed under the experimental conditions.

Figure 2 shows the yields of the inclusion crystals formed from benzene, ethylbenzene, and their 1:1 mixture as a function of the guest/host molar ratio in the feed solution. The yield was calculated as a ratio of the amount of CA crystallized to that in the feed solution. In the case of benzene as guest, the formation of the crystals occurs abruptly when the guest/host ratio exceeds *ca.* 10, and the yield reaches over 70%. A further increase in the ratio does not affect the yield. On the other hand, the yield of CA-ethylbenzene reaches 84%. This higher yield would be ascribed to a lower solubility of CA in ethylbenzene than in benzene, though a higher

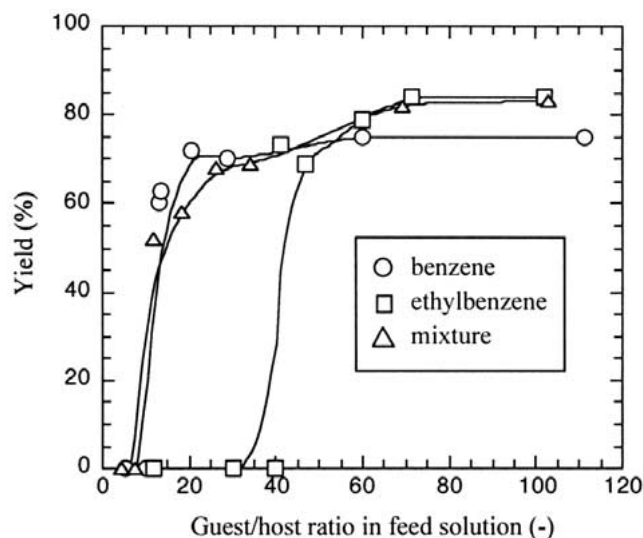


Figure 2. Relationship between yield of CA inclusion crystal and guest/host molar ratio in feed solution.

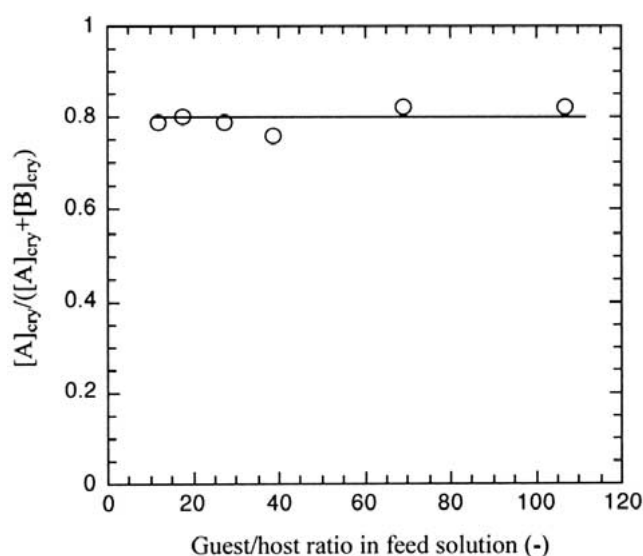


Figure 3. Relationship between selectivity of benzene in CA inclusion crystal and guest/host molar ratio in feed solution.

guest/host ratio (>*ca.* 40) is required to obtain the crystal. Furthermore, the 1:1 mixture of the present two guests is found to give a yield curve similar to the case of benzene.

Figure 3 shows the molar ratio of the two guests (A: benzene, B: ethylbenzene) in the CA inclusion crystals, $[A]_{cry}/([A]_{cry} + [B]_{cry})$, made from 1:1 guest mixtures against the guest/host molar ratio in the feed solution. The subscript *cry* denotes the crystal phase. All the values locate almost on a single straight line of 0.8, indicating that benzene is preferentially included over ethylbenzene irrespective of the guest/host ratio. This behavior agrees with the similarity of the yield curves for the two inclusion crystals from benzene and the mixture, as shown in Figure 2. The invariable selectivity also means that a constant amount of ethylbenzene is incorporated in the ternary crystal, though the inclusion crystal of CA-ethylbenzene is not formed when the guest/host ratio is below 40 (see Figure 2). It seems that the presence of benzene encourages ethylbenzene to be in-

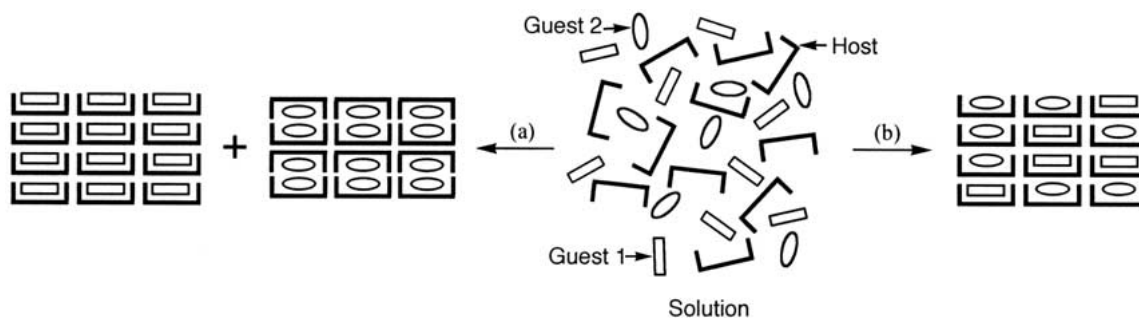


Figure 4. Schematic representation of formation of inclusion crystals from two guests.

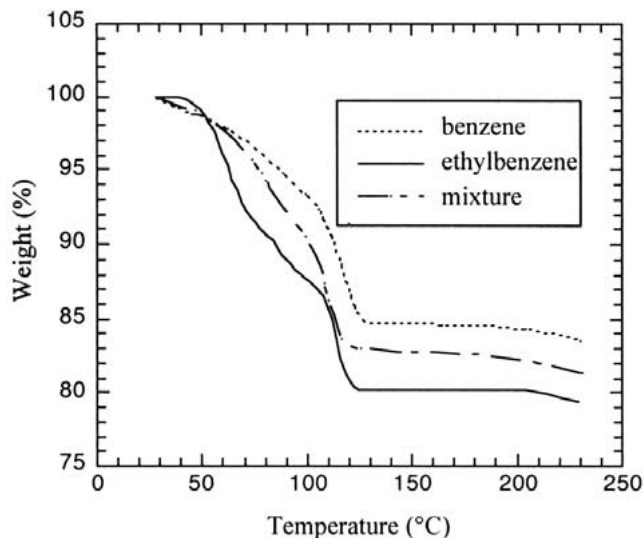


Figure 5. TG diagrams of CA inclusion crystals obtained from benzene, ethylbenzene, and their 1:1 mixture.

cluded. The above results imply that the resulting crystals are not the mixture of two distinct crystals containing either of the two guests (Figure 4a). Consequently, the resulting crystal consists of the two guests mixed in a single host framework, as shown in Figure 4b.

Figure 5 shows the results for thermogravimetric analysis for the inclusion crystals obtained from benzene, ethylbenzene, and the 1:1 mixture solution. Regardless of a difference in boiling point of benzene (80 °C) and ethylbenzene (136 °C), the guest molecules are released completely up to 120 °C. The observed weight loss and calculated one in parenthesis are 15% (16%) for CA·benzene, 20% (21%) for CA·ethylbenzene, and 17% (17%) for CA·mixed guests with 8:2 molar ratios. We have already reported that each guest gives an inclusion crystal at 1:1 host/guest molar ratio by means of X-ray crystallography [7], and found that the calculated values agree with the observed ones. The shape of TG curve for the crystal obtained from 1:1 mixture is similar to that from benzene. This similarity is ascribed to the fact that the ternary crystal has the same host framework with CA·benzene, which will be discussed later.

Relationship between selectivity and host framework

Inclusion crystals were obtained from mixed guest solutions having various concentrations of benzene and ethylbenzene.

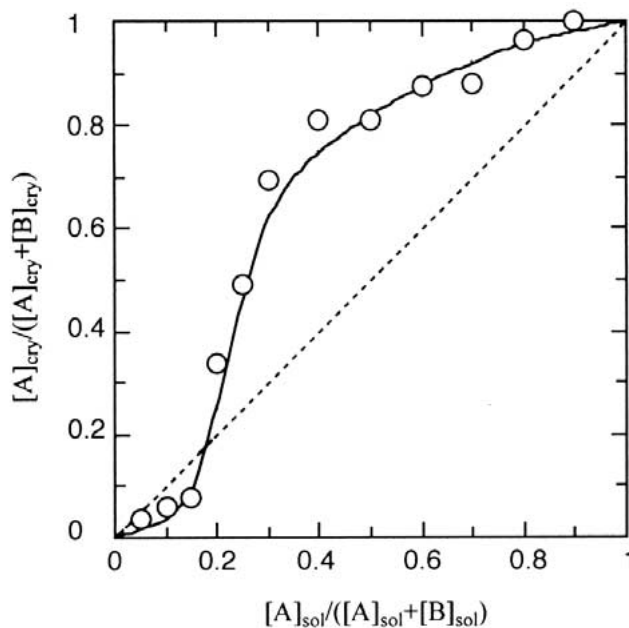


Figure 6. Relationship between molar ratio of two guests in CA crystal and in residual solution.

A total amount of the two guests was set to be 20 mmol (guest/host = 62.5) to provide the similar solubility of CA inclusion crystals (see Figure 2). Figure 6 shows the molar ratio of benzene in the crystals, $[A]_{cry}/([A]_{cry} + [B]_{cry})$, against that in the residual solution, $[A]_{sol}/([A]_{sol} + [B]_{sol})$, where the subscripts *cry* and *sol* denote the crystal and solution phase, respectively. The ratio in the crystal locates above the diagonal line for the most part, indicating that benzene is preferentially incorporated in the CA crystal over ethylbenzene. However, the guest preference reverses when the ratio in the solution is less than 0.2.

As reported earlier [11], the preference of benzene is attributed to difference in the host framework types, as shown in Figure 7. The structures of CA·benzene and CA·ethylbenzene crystals can be classified into sub-types of α -*gauche* and β -*trans* host frameworks, respectively, depending on the difference in interdigitation manners of methyl groups in the lipophilic faces (α and β types) and in steroidal side chain conformations (*gauche* and *trans* types) [9]. Shape fit between the host cavity and guest molecule plays an important role for selectivity under competitive co-crystallization. The host cavity of α -*gauche* host framework

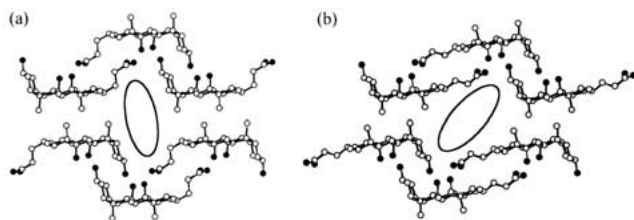


Figure 7. Crystal structures; (a) CA-benzene in α -*gauche* type, (b) CA-ethylbenzene in β -*trans* type. Carbon and oxygen atoms are represented by open and filled circle, respectively. Guest molecules are omitted and ellipses represent inclusion spaces.

has a square groove that is appropriate for a phenyl ring of a guest molecule. This leads to dominant formation of α -*gauche* host framework and the preferential incorporation of the benzene molecule.

In order to clarify the reversal in selectivity for the two guests (see Figure 6), we investigated host framework types of the resulting crystals containing benzene and ethylbenzene at various molar ratios. Figure 8 shows X-ray powder diffraction patterns of CA-benzene (a), CA-ethylbenzene (e), and CA crystals obtained from the benzene/ethylbenzene mixtures at the ratio of 3:7 (b), 2:8 (c), and 1:9 (d); the molar ratios of benzene included in the crystals at these points were 0.70, 0.34, and 0.06, respectively. The diffraction patterns of CA crystals from the mixtures (b) and (c) are very similar to that of CA-benzene (a), i.e. the mixture of the two guests is incorporated in α -*gauche* type cavity. It is to be noted that, the host framework of α -*gauche* type is dominantly formed owing to the high affinity for benzene, even though the amount of benzene is smaller than that of ethylbenzene in the crystal obtained from the guest mixtures at the ratio of 2:8. On the other hand, the pattern of CA crystal from the mixture (d) is similar to that of CA-ethylbenzene (e), indicating the formation of β -*trans* host frameworks. This behavior agrees with the reversal in selectivity for the two guests. In conclusion, the formation of a certain host framework is dependent on the amount of each guest in the system, and is dominant factor for the selective incorporation.

Conclusion

We demonstrated selective incorporation of two aromatic compounds, benzene and ethylbenzene, into the inclusion crystal of CA. The selectivity varies depending on the ratio of the two guests but not on the guest/host ratio in the feed solution. The guest preferentially incorporated is dependent on the formation of a certain host framework, indicating that separation behavior is closely related with structural flexibility of a host framework. As in the case of benzene/ethylbenzene as seen in Figure 6, the reversal in selectivity would be expected in all the competitive co-crystallization for two guests that give different types of the host frameworks. Since CA has more than twelve host frameworks dependent on guest compounds, we believe that CA is a promising and flexible separation medium applicable for a wide variety of organic mixtures.

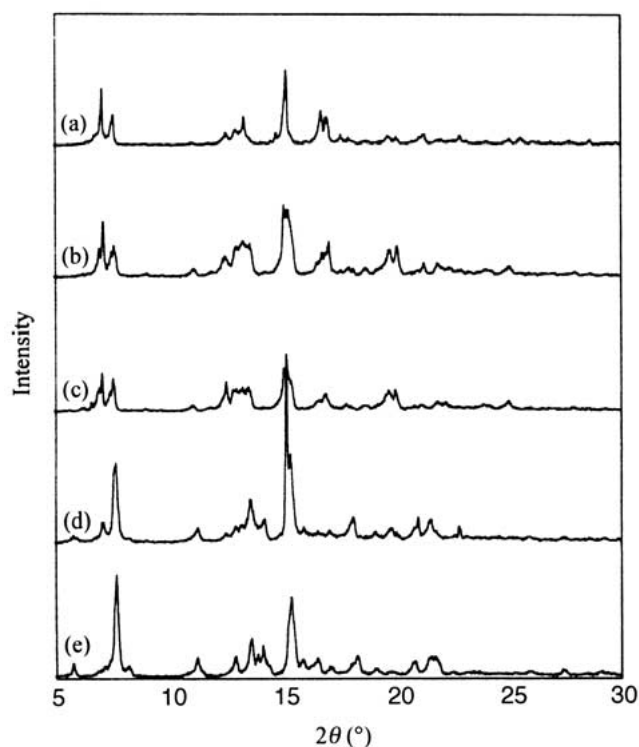


Figure 8. X-ray diffraction patterns of CA inclusion crystals obtained from (a) benzene, (b) 3:7 benzene/ethylbenzene mixture, (c) 2:8 mixture, (d) 1:9 mixture, and (e) ethylbenzene.

Acknowledgements

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