Molecular States of 2-Naphthoic Acid in Solid Dispersions with Porous Crystalline Cellulose, as Investigated by Fluorescence Spectroscopy

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2-Naphthoic acid (2-NPA) crystals were mixed with porous crystalline cellulose (PCC). After a heat-treatment of the mixture, no diffraction peaks due to 2-NPA crystals were observed on the XRD pattern. Fluorescence spectral data indicated that the molecular state of 2-NPA was modified by heating with PCC. The results of a fluorescence analysis also seemed to support the molecular state change of 2-NPA. When 2-NPA formed an inclusion complex by co-grinding with γ-cyclodextrin (γ-CD), the emission spectra of 2-NPA in this sample was similar to that in the heat-treated sample with PCC. Since γ-CD could be assumed to include two naphthyl moieties into the cavity, the fluorescence properties of 2-NPA after a heat treatment with PCC could be characterized by the excimer of 2-NPA on the PCC surface.

In order to improve the dissolution profile of water-insoluble drugs, solid dispersion systems have often been prepared.[1,2] Porous materials are useful excipients for solid dispersion, because of their huge specific surface area, reflecting their porous structure. Porous crystalline cellulose (PCC), characterized by a porous structure, is derived from microcrystalline cellulose (MCC). Since MCC is a well-known pharmaceutical excipient for tablet preparation, porous crystalline cellulose seems to be applicable for practical use. It has been reported that drug molecules became an amorphous state when they were stored with porous materials by the adsorption of drug molecules onto the surface of the porous materials.[3] An improvement on the dissolution rate of the drugs from the solid dispersion with porous materials was also reported.[4]

Fluorescence measurement is utilized as one of the effective approaches to clarify the structure and dynamics of biomolecules in solution[5] and to study molecular interactions, such as a surfactant–polymer interaction in solution.[6] Recently, fluorescence analysis has also been applied to solid systems in order to study molecular interactions, such as inclusion phenomena of cyclodextrin and guest molecules,[7,8] and to study physicochemical changes of medicinal with adsorbate on solid surfaces.[9,10] These studies proved that fluorescence could be useful to investigate the molecular state in solid dispersions.

In this study, we performed a solid-state fluorescence measurement to investigate the molecular state of drugs adsorbed on a PCC surface. 2-Naphthoic acid was chosen as a model compound because the naphthyl groups showed excimer fluorescence when the naphthalene rings were overlapped in a parallel conformation;[11] the carboxyl group showed a peak shift of the carbonyl stretching band in IR spectra when interacted with other functional groups.[12]

Experimental

Materials: 2-Naphthoic acid (2-NPA; Nacalai Tesque, Kyoto, Japan) of reagent grade was used without further purification. Porous crystalline cellulose (PCC; mode pore diameter of 40 Å and specific surface area of 87.2 m^2) was supplied by Asahi Chemical Industrial Co., Ltd., Japan. Microcrystalline cellulose (Avicel PH101; specific surface area of approximately 1.0 m^2) was purchased from Asahi Chemical Industrial Co., Ltd., Japan. These additives were used after drying under reduced pressure at 110 °C for 3 h. β-Cyclodextrin (β-CD; Mercian, Tokyo, Japan) and γ-Cyclodextrin (γ-CD; Mercian, Tokyo, Japan) were used after drying under reduced pressure at 120 °C for 3 h.

Sample Preparation: Simple mixtures were prepared by blending of 2-NPA and additives with predetermined mixing ratios in a glass vial for 1 min. A simple mixture (about 250 mg) was sealed in a glass ampoule (2 mL) and then heated at 100 °C for 3 h to prepare a heated sample. Ground mixtures were prepared by grinding a simple mixture (3.0 g) of 2-NPA and CDs by means of a vibrational mill (CMT TI-200, Tochigi, Japan) made of alumina.

Powder X-Ray Diffractometry: Powder X-ray diffraction was performed using a Rigakudenki Miniflex diffractometer (Tokyo, Japan). The measurement conditions were as follows: target, Cu; filter, Ni; voltage, 30 kV; current, 15 mA; count range, 2000 cps; scanning speed, 2 ° min^{-1}.

Fourior Transform Infrared (FT-IR) Spectroscopy: Fourior-transform infrared spectra were measured by the KBr disc method at a resolution of 2 cm^{-1} for 32 scans using a JASCO 230 FT-IR spectrophotometer (Tokyo, Japan).

Thermal Analysis: A thermogravimetry-differential thermal analysis (TG-DTA) was carried out using a MAC Science TG-DTA 2000 (Japan) at a heating rate of 5 °C min^{-1} under a nitrogen gas flow.

Fluorescence Spectroscopy: An FP-770F fluorescent spectrometer (Japan Spectroscopy Co., Ltd., Tokyo, Japan) was used for stationary fluorescence spectroscopy. Powder samples were filled into a front-face reflectance cell (FP-1060).

Determination of the Fluorescence Lifetime and Time-Resolved Fluorescence Spectra: Measurements of fluorescence decay curves were carried out by a nanosecond time-resolved single-photon counter with a pulse width of 1.5 ns (Horiba NAES-770, Tokyo, Japan). The exciting pulse and emission response functions were measured simultaneously, and to compute the decay parameters (up to three components) following deconvolution of the excitation pulse using a non-linear least-squares fitting. The best fit for an observed fluorescence decay curves was obtained by minimizing the value of χ^2.

Results and Discussion

1. Molecular States of 2-NPA Molecules in the Mixture with Cellulose. Figure 1 shows the powder X-ray diffraction patterns of simple mixtures and heated samples of 2-NPA with PCC or MCC. The diffractogram of simple mixtures were a superimposed pattern of the two components, 2-NPA and each additive. The diffraction peaks of 2-NPA crystals disappeared after sealed heating with PCC, indicating the disappearance of an ordered arrangement of 2-NPA molecules in the crystal, while the X-ray diffraction peaks of 2-NPA crystals remained after heating with MCC.

Figure 2 shows the changes in the fluorescence emission spectra when the excitation wavelength was fixed at 262.7 nm. The emission spectra observed in the 2-NPA–MCC system was identical to that of 2-NPA crystals, while the emission peak of 2-NPA in the simple mixture with PCC was observed at a longer wavelength than that of the crystals. The emission spectrum of the heated sample with PCC had a maximum around 410 nm, and was clearly different from that of 2-NPA crystals.

Fluorescence-decay kinetics was investigated at excitation and observation wavelengths of 262.7 and 376.0 nm, respec-

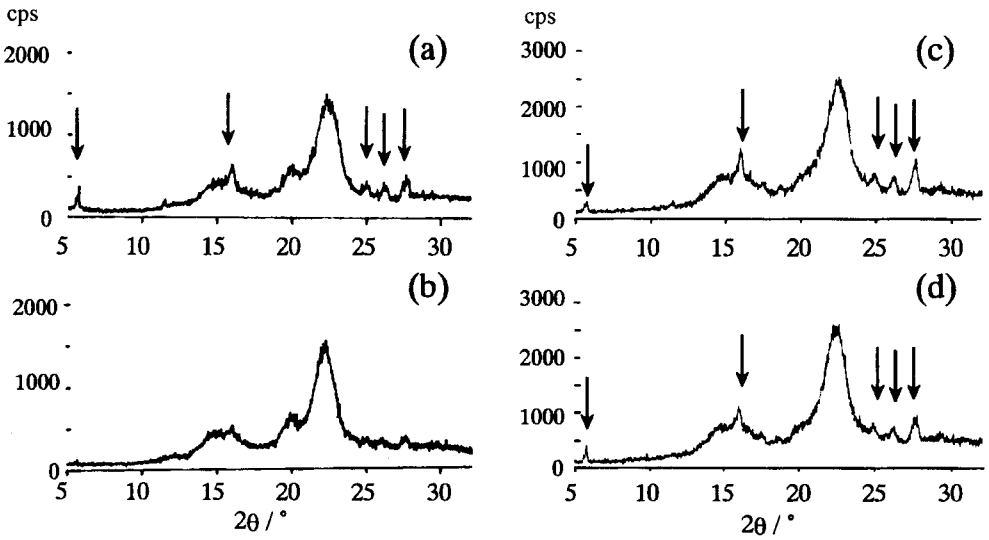

Fig. 1. Powder X-ray diffraction patterns of 10% 2-naphthoic acid (2-NPA)–cellulose systems; (a) simple mixture of 10% 2-NPA and 90% PCC, (b) sealed heated sample of *a*, (c) simple mixture of 10% 2-NPA and 90% MCC, (d) sealed heated sample of *c*.

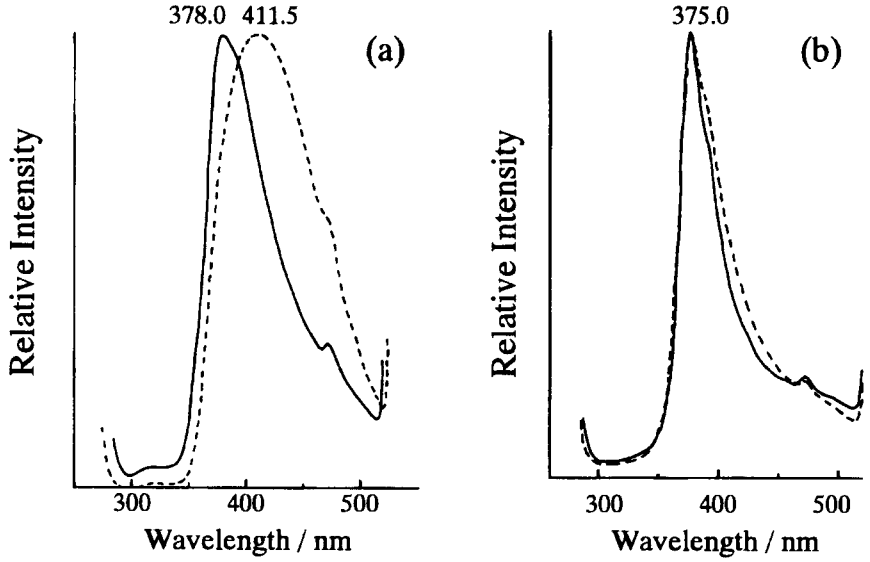

Fig. 2. Changes in the solid-state fluorescence spectra of 10% 2-NPA–cellulose system, λ_{ex} = 262.7 nm, (a) 2-NPA–PCC system, (b) 2-NPA–MCC system; simple mixture (—); sealed heated at 100 °C for 3 h (- - -).

tively. The lifetime and relative quantum yield are listed in Table 1. The component of short lifetime (τ_1) could be due to stray light.[13] Since there exists a distribution of fluorescence lifetimes for the probe molecules in the solid-state, it may be difficult to assign a calculated value of lifetime to a fixed physical species of the molecule. Nevertheless, the results of a lifetime analysis seem to suggest some physicochemical changes of 2-NPA during the process. Therefore, we attempted to compare the lifetime data to help our discussion concerning the adsorption mechanism.

As shown in Table 1, both the fluorescence lifetimes and the relative quantum yields of 2-NPA observed in the MCC system were similar to those observed in 2-NPA crystals. On the other hand, in the simple mixture and the heated sample with PCC, a longer lifetime component of 2-NPA was observed compared to that of 2-NPA crystals. It was noteworthy that the component having a longer lifetime (11.9 ns) was observed after 1 min of mixing with PCC. The lifetime of the longest component was changed by the heat treatment (15.8 ns), and the emission spectrum in this system was quite different from that in the crystal. These results showed that 2-NPA molecules changed their molecular states in the 2-NPA–PCC system.

2. Molecular State Changes of 2-NPA by Co-grinding with Cyclodextrins. Cyclodextrins form inclusion complexes with many organic compounds through the accommodation of a guest molecule into the cavity.[14] Since the inclusion compound with CD showed unique characteristics in fluorescence studies compared to the crystals, we performed a co-grinding procedure of 2-NPA and CD. Figure 3 shows the powder X-ray diffraction patterns of 2-NPA crystals, an equimolar simple mixture and ground mixtures with β-CD. The diffraction-peak intensities of 2-NPA and β-CD crystals decreased with the duration of the grinding time, and a halo pattern was found for the 30 min ground mixture, indicating an amorphous state of 2-NPA and β-CD. Figure 4 shows the IR spectra of 2-NPA with β-CD systems (1 : 1 molar ratio). The infrared band observed at 1685 cm^{-1} in the spectrum of 2-NPA crystals was assigned to the carbonyl stretching vibration of 2-NPA, which shifted to a higher wave number with increasing grinding time. The carbonyl stretching band of 2-NPA in the 30 min ground mixture was observed at 1700 cm^{-1}, which was at a 15 cm^{-1} higher position compared

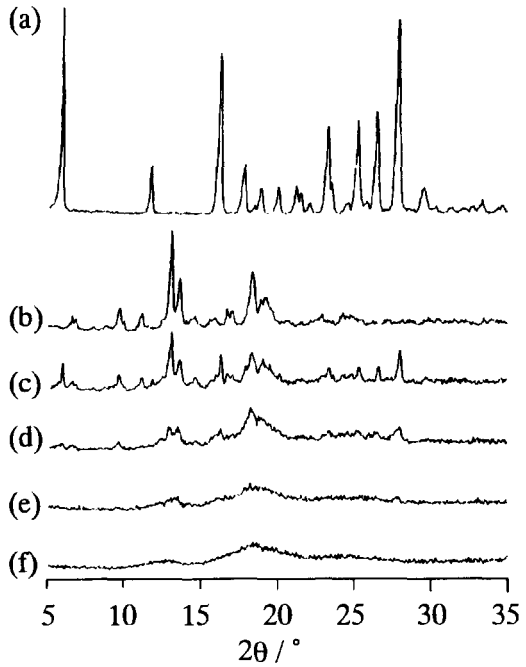

Fig. 3. Changes in powder X-ray diffraction patterns of 2-NPA–β-CD system (molar ratio of 2-NPA : β-CD = 1 : 1). (a) 2-NPA crystals; (b) β-CD crystals; (c) simple mixture; (d) ground for 5 min; (e) ground for 10 min; (f) ground for 30 min.

with that of 2-NPA crystals. The thermogravimetry curves shown in Fig. 5 illustrate the sublimation behavior of 2-NPA from the simple mixture and the ground mixtures with β-CD. In the TG curves from 120 to 200 °C, the weight loss from the ground mixture was significantly decreased with the duration of the grinding time. This suppression of 2-NPA sublimation was considered to have resulted from the formation of an inclusion complex between 2-NPA and β-CD.

Harata reported that, in the crystal structure of the 1 : 1 complex between heptakis (2,6-di-O-methyl)-β-cyclodextrin (DM-β-CD) and 2-NPA, the naphthalene moiety was included in the host cavity.[16] Since the cavity size of β-CD was almost the same as that of DM-β-CD, we may assume that β-CD also forms a 1 : 1 complex with 2-NPA. Complexation during the grinding process does not necessarily proceed quantitatively; therefore, an excess of β-CD was

Table 1. Changes of Fluorescence Lifetime (τ) and Relative Quantum Yield (Q) of 10% 2-NPA–Cellulose System

λ_{ex} = 262.7 nm, λ_{obs} = 376.0 nm, Sealed heating (SH) was carried out at 100 °C for 3 h.

	τ_1/ns	Q_1/%	τ_2/ns	Q_2/%	τ_3/ns	Q_3/%	χ^2
2-NPA crystals	0.216	23.9	3.12	53.2	7.89	22.9	1.03
2-NPA–MCC system							
Simple mixture	0.093	29.7	3.02	41.4	7.98	28.9	1.41
SH sample	0.317	23.4	3.10	43.9	8.76	32.7	1.12
2-NPA–PCC system							
Simple mixture	0.279	27.4	3.55	34.6	11.9	38.0	1.06
SH sample	0.208	24.3	4.76	31.6	15.8	44.0	1.09

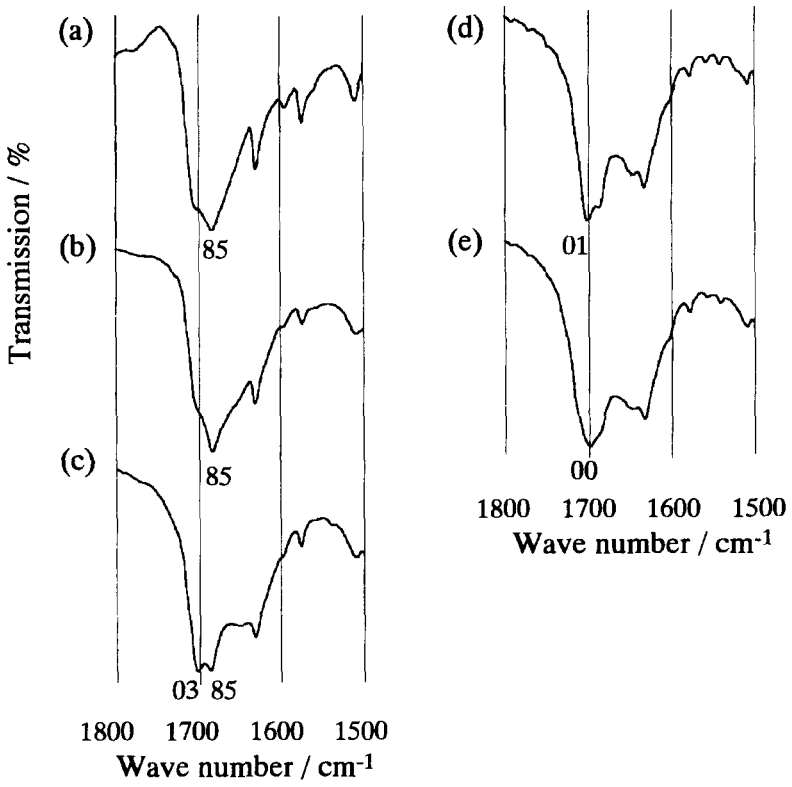

Fig. 4. IR spectra of the ground mixtures of 2-NPA–β-CD system (molar ratio of 2-NPA : β-CD = 1 : 1). (a) 2-NPA crystals; (b) simple mixture; (c) ground for 5 min; (d) ground for 10 min; (e) ground for 30 min.

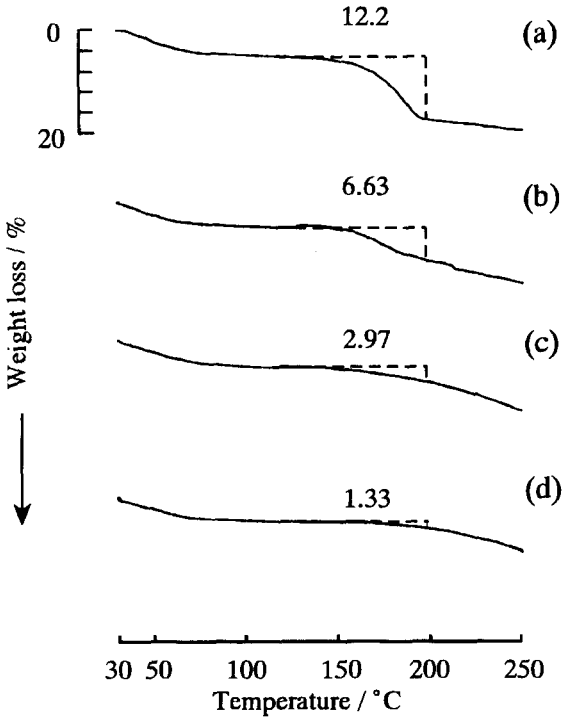

Fig. 5. Effect of grinding time on TG curves of 2-NPA–β-CD system (molar ratio of 2-NPA : β-CD = 1 : 1), (a) simple mixture, (b) ground for 5 min, (c) ground for 10 min, (d) ground for 30 min.

loaded for all portions of 2-NPA to be accommodated into the cavity. To investigate the fluorescence properties of included 2-NPA, co-grinding for the 2-NPA–β-CD system was carried out at a molar ratio of 1 : 2.

Changes in the fluorescence emission spectra of 2-NPA by grinding with β-CD are shown in Fig. 6. The increase in the grinding time resulted in a slight shift of the emission maximum from 376.0 to 378.0 nm. This was quite similar to the emission spectra of the simple mixture with PCC. In Table 2, a component having a long lifetime of more than 12.0 ns was observed only in the ground mixtures, and the relative quantum yield of this component increased with the duration of grinding. This lifetime of 2-NPA was similar to the long-lifetime component in the simple mixture with PCC (11.9 ns). From the results, 2-naphthoic acid molecules in the simple mixture with PCC was considered to exist as a monomer state.

Figure 7 exhibits the changes in the fluorescence emission spectra of the 2-NPA–γ-CD mixture by grinding (molar ratio of 2-NPA : γ-CD = 2 : 1). After 30 min of grinding, a broad emission peak was observed at a longer wavelength of 403.5 nm. The notable peak shift of 2-NPA observed in the γ-CD system was similar to that observed in the heated sample with PCC.

Figure 8 shows the time-resolved fluorescence spectra of 2-NPA in the 30 min ground mixture with γ-CD. At the early stage of the time interval (0—6 ns), the emission-peak position was coincident with the fluorescence spectrum of crystalline 2-NPA. This indicates that a short-lifetime com-

Table 2. Changes of Fluorescence Lifetime (τ) and Relative Quantum Yield (Q) of 2-NPA–β-CD System
(molar ratio of 2-NPA : β-CD = 1 : 2, λ_{ex} = 262.7 nm, λ_{obs} = 376.0 nm)

	τ_1/ns	Q_1/%	τ_2/ns	Q_2/%	τ_3/ns	Q_3/%	χ^2
2-NPA crystals	0.216	23.9	3.12	53.2	7.89	22.9	1.03
Simple mixture	0.284	26.1	2.95	43.2	7.94	30.6	1.41
Ground for 5 min			4.53	26.3	12.6	73.7	1.32
Ground for 10 min			4.61	25.7	12.7	74.3	1.13
Ground for 30 min			4.66	22.7	12.1	77.3	1.39

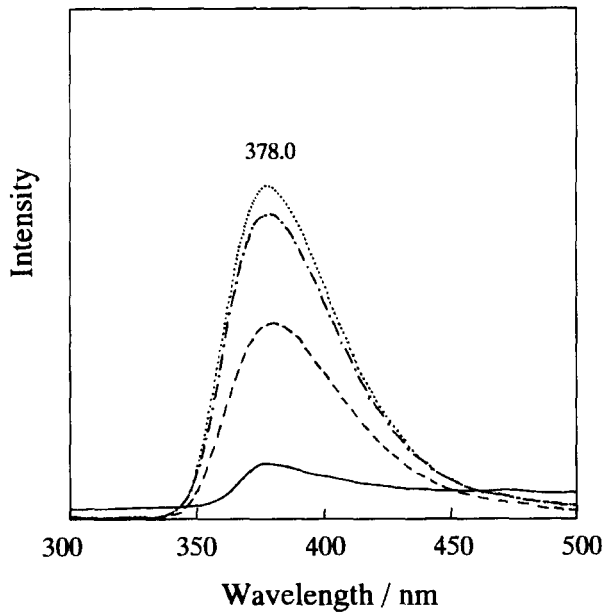

Fig. 6. Solid-state emission spectra of 2-NPA–β-CD system (molar ratio of 2-NPA : β-CD = 1 : 2), λ_{ex} = 262.7 nm. Simple mixture (—); ground for 5 min (— —); ground for 10 min (— - —); ground for 30 min (-------).

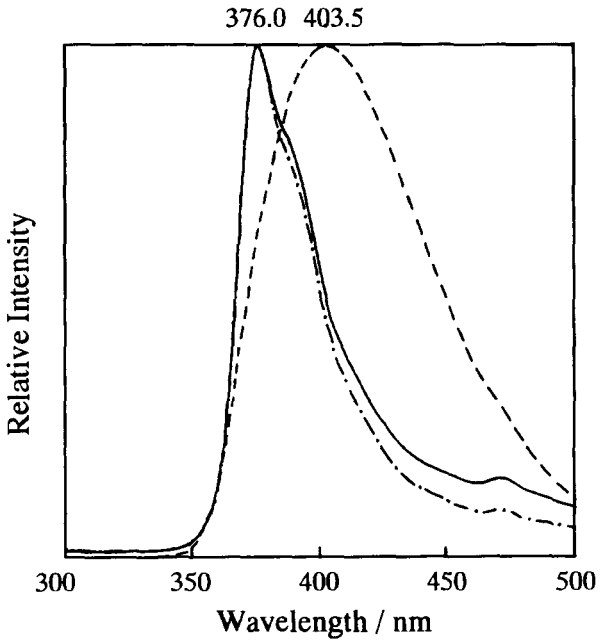

Fig. 7. Solid-state emission spectra of 2-NPA–γ-CD system (molar ratio of 2-NPA : γ-CD = 2 : 1), λ_{ex} = 262.7 nm. 2-NPA crystals (—); simple mixture (— - —); ground for 30 min (— —).

ponent arose from the molecules in 2-NPA crystals. The peak maximal position gradually shifted to a longer wavelength with the time interval, suggesting that the long-lifetime component did not arise from 2-NPA molecules in the crystals. The changes in the fluorescence spectra indicate that there were at least two different species characterized by different fluorescence lifetimes in the ground mixture, and that their contribution to the entire fluorescence emission changed with time. With regard to the 2-NPA–γ-CD system, the lifetimes and relative quantum yields are listed in Table 3. A fluorescent component having a long lifetime (τ = 19.8 ns) was found in the 30 min ground mixture.

It was reported that naphthalene showed excimer emission when the two naphthalene rings overlapped in a parallel conformation, and that compounds having the naphthyl moiety were also able to form the excimer, e.g., naphthalenophanes,[11] cyclodextrin derivative bearing two naphthyl molecules.[17,18] When the two naphthyl moieties had the overlapped geometry, an emission peak due to the excimer fluorescence appeared around 410 nm. It was also reported that the naphthalene excimer was characterized by the longer fluorescence lifetime than the naphthalene monomer.[10,19] In this study, the excimer emission of the 2-NPA molecule was observed in the γ-CD ground mixture, while only monomer emission was observed in the β-CD

Table 3. Changes of Fluorescence Lifetime (τ) and Relative Quantum Yield (Q) of 2-NPA–γ-CD System
(molar ratio of 2-NPA : γ-CD = 2 : 1, λ_{ex} = 262.7 nm, λ_{obs} = 376.0 nm)

	τ_1/ns	Q_1/%	τ_2/ns	Q_2/%	τ_3/ns	Q_3/%	χ^2
2-NPA crystals	0.216	23.9	3.12	53.2	7.89	22.9	1.03
Simple mixture	0.093	29.7	3.02	41.4	7.98	28.9	1.40
Ground for 30 min	0.147	12.2	4.88	32.0	19.8	55.8	1.15

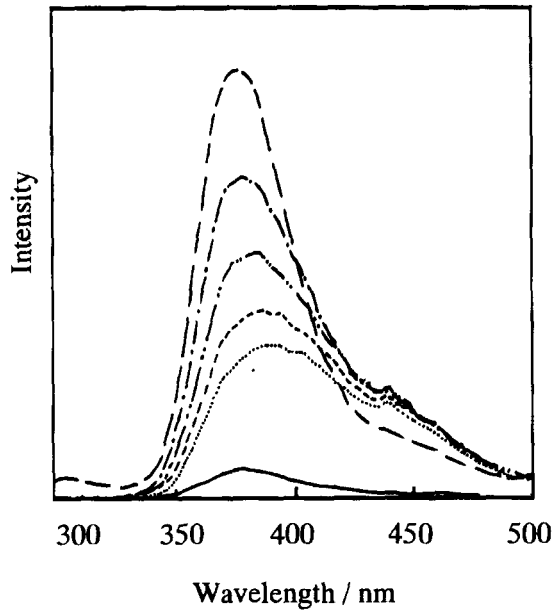

Fig. 8. Time-resolved fluorescence spectra of 2-NPA coground with γ-CD for 30 min (molar ratio of 2-NPA : γ-CD = 2 : 1), λ_{ex} = 262.7 nm. Time intervals for determining the spectrum: 0—3 ns (—); 3—6 ns (— —); 6—9 ns (–·–); 9—12 ns (–··–); 12—15 ns (- - -); 15—18 ns (····).

ground mixture. Taking account of the cavity sizes of cyclodextrins, γ-cyclodextrin seemed to be able to accommodate two naphthyl moieties into the cavity, while the β-CD cavity was not large enough. The broad peak at 403.5 nm observed in the 2-NPA–γ-CD ground mixture, therefore, could be due to excimer emission that arose from two overlapped 2-NPA molecules in the γ-CD cavity.

We obtained similar results of the fluorescence studies in heated samples with PCC and in the co-ground sample with γ-CD. It could be reasonable to assume that both systems showed the excimer emission of 2-NPA around 410 nm in the solid-state fluorescence spectra, and presented a lifetime component of more than 15.0 ns in the lifetime analysis. The mode pore diameter of PCC was estimated to be as small as 40 Å. Because of the small pore, the neighbouring 2-NPA molecules in the pore could be aligned to have a face-to-face configuration, showing excimer emission.

Conclusions

When 2-NPA was adsorbed onto the PCC surface by heating, physicochemical changes of 2-NPA were observed on the fluorescence spectra and fluorescence decay curves. After heating with PCC, 2-NPA molecules show excimer emission and a longer fluorescence lifetime than its crystal. It was concluded from solid-state fluorescence spectroscopy that the molecular state of 2-NPA could be easily varied in the presence of PCC.

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