Encapsulation and Release Characteristics of Carbon Dioxide in α-Cyclodextrin

TZE-LOON NEOH, HIDEFUMI YOSHII* and TAKESHI FURUTA
Department of Biotechnology, Tottori University, 4-101 Koyamacho-minami, 680-8552, Tottori, Japan

Abstract

Encapsulation and release of carbon dioxide (CO_2) into and from α-cyclodextrin (α-CD) was studied. Initial moisture content of α-CD and CO_2 pressure were found to have affected the encapsulation behavior. The increase of CO_2 pressure has constantly accelerated the encapsulation rates and increased the maximum inclusion ratio, whereas the increase of initial moisture content showed no consistent effect. The saturated α-CD solution produced the inclusion complex crystal of similar inclusion ratio to solid α-CD. The release characteristics of inclusion complexes were also monitored at various relative humidities at 25 °C. Predominantly, increase in storage humidity accelerated the release of CO_2. The inclusion complex crystal prepared from saturated α-CD solution showed the most stable release characteristic at all storage humidities investigated. The encapsulation and the release characteristics were analyzed using the first-order reaction equation and the Avrami's equation respectively, in order to estimate the rate processes of encapsulation and release. The FT-IR spectra of inclusion complexes presented an absorption band at wavenumber around 2338 cm^{-1}, indicating CO_2 molecules resided inside the α-CD cavities in gaseous state rather than being bound to the hydroxyl groups of α-CD. Powder X-ray diffractometry was carried out to investigate the crystal lattice structure of α-CD and inclusion complexes. Scanning electron microscopy was also performed for morphological examination.

Introduction

After a century of continuous research and development, cyclodextrins (CDs) have gained certain recognition of its importance in various fields including foods, pharmaceuticals, agricultural, analytical, cosmetics, personal care, etc. Their applications are mainly intended for the entrapment of smaller molecules, stabilization of reactive intermediates, catalysis through encapsulation and as potential molecular transport and drug delivery device [1]. In food related applications, flavor compounds are being encapsulated into CDs for better retention and protection from various possible means of deterioration, as well as for controlled delivery. In pharmaceutical field, publications were made based on study on solubility enhancement of poorly water-soluble drugs [2–4].

CDs are also being utilized as carriers for controlled release of particular compounds entrapped within the cavities. Particularly in pharmaceutical industry, controlled release systems are desirable to give optimized efficacy, safety and convenience because they can be designed to deliver a drug at a specified rate, for a specific period of time and even at a desired location [5]. Performance of CDs as carrier in controlled release systems can be evaluated by the release characteristics of inclusion complexes at various parameters of interest. Application of a specific inclusion complex is to a great extent dependent on its release characteristic. In the 1950s, feasibility of molecular encapsulation of gases within α-CD cavity was first highlighted by Cramer and Henglein [6]. Aqueous α-CD was reported to include gaseous hydrocarbons (methane, ethylene, ethane, propane), Cl_2, Kr, Xe, O_2 and CO_2 to form clathrates. A number of synthetic compounds (cryptophanes, carcerands, calixarenes) were also being discussed over their gas encapsulation ability in a recent review by Rudkevich and Leontiev [1]. Purification of gases based on the selective adsorption of components of gas mixture onto an adsorbent material was discussed by Atwood *et al.* [7]. The high affinity of CO_2 for complex formation is utilized for the isolation of α-CD through the precipitation of a gas inclusion complex [8]. Encapsulation of 1-methylcyclopropene, an ethylene response inhibitor, into α-CD may also be considered one of the worth noting examples. This inclusion complex has been developed, patented by Daly and Kourelis [9] and launched into some regional markets by Rohm and

* Author for correspondence. E-mail: yoshii@bio.tottori-u.ac.jp

Haas Company. However, the encapsulation behavior and release characteristic of this particular inclusion complex still remain available for exploration.

CO_2, a non-polar gas owing to its symmetrical arrangement, bears claims of several beneficial physiological effects namely blood vessel dilation, blood circulation improvement and activation of gastrointestinal movement. In view of that, encapsulation of CO_2 with CDs was earlier patented in Japan in 1987 anticipating its uses in cosmetics, cleansing and personal care products [10]. A bath salt formulation incorporating CO_2–α-CD complex was patented by Kao Corporation Japan [11].

In this research, microencapsulation of CO_2 into α-CD was studied with respect to encapsulation behavior and release characteristic. In encapsulation, focus was placed on the effect of initial moisture content of α-CD and the CO_2 pressure employed. Encapsulation behavior was assessed in terms of final CO_2 inclusion ratio and encapsulation rate. Release characteristic was studied with respect to the CO_2 retention and CO_2 release rate, focusing on the effects of storage humidity and initial moisture content. Fourier transform infrared (FT-IR) spectrometer was utilized to confirm the inclusion of CO_2 into the molecular cavity of α-CD. Changes in crystalline structure of α-CD after CO_2 encapsulation was determined by powder X-ray diffractometry (PXRD). Morphology of α-CD crystal and inclusion complexes were also observed with scanning electron microscope (SEM).

Experimental

Materials

The α-CD with 99% minimum purity was purchased from Ensuiko Sugar Refining Co., Ltd. (Tokyo, Japan). The α-CD powder was dried under vacuum at 90 °C for 24 h before use. The gaseous CO_2 was purchased from Iwatani International Corporation (Osaka, Japan). The rest of the chemicals were purchased from Wako Pure Chemical Industries, Ltd. (Osaka, Japan). Acetone was subjected to dehydration before use. Distilled water was used throughout the entire experiment. Unless otherwise stated, the chemicals used in the experiments were of reagent grade.

Preparation of CO_2–α-CD inclusion complex

The encapsulation was carried out under CO_2 pressure using High Pressure Microreactor-MMJ 200 (OM Lab-Tech Co., Ltd., Tochigi, Japan) with a stainless steel vessel (38.5 mm in diameter and 197 mm in depth) that is able to withstand pressure up to 19.6 MPa. Encapsulation of CO_2 was done in two conditions of α-CD: solid state at different initial moisture contents (2, 10 and 30 wt%) and aqueous phase (saturated α-CD solution at 25 °C). For encapsulation into solid α-CD,

approximately 10 g of moistened α-CD was inserted into the vessel. Subsequently, CO_2 was filled under pressure of either 1 or 3 MPa into the vessel. For encapsulation in saturated solution, approximately 150 ml sample was added into the vessel which was then pressurized with CO_2 at 3 MPa. After encapsulation, crystals from aqueous solutions were separated from liquid by vacuum filtration. The collected inclusion complex was washed with cold water and then dehydrated acetone before being filtered again.

Release experiment

The release characteristics of inclusion complexes were carried out at constant temperature and relative humidity. Inclusion complex of ca 0.1 g was inserted into 30-ml Erlenmeyer flasks, which were stored in a desiccator with constant relative humidity at 25 °C. The relative humidity was controlled independently at 5, 33 and 75% using silica gel, saturated salt solutions of magnesium chloride and sodium chloride, respectively [12]. The desiccators were placed in an oven (Masuda SA460, Masuda Corp., Osaka, Japan) adjusted at 25 °C for at least 24 h before use. A thermo recorder (TR-72U, T&D Corp., Nagano, Japan) was place inside the desiccator to monitor the relative humidity and temperature of desired values. The relative humidities were obtained at the intended values within a range of ±5% RH. In the release experiment, the flasks containing inclusion complex were placed in desiccators with different relative humidities over a storage period of 1 week (168 h). Three Erlenmeyer flasks were withdrawn from the desiccator after each storage time of 1, 2, 3, 4, 24, 96 and 168 h for measurement of CO_2 retained. The CO_2 retention in the complex powder was defined as the ratio of CO_2 remained after a particular storage time to the initial amount.

Quantification of CO_2 in the complex powder

The quantitative analysis of CO_2 encapsulated in α-CD was conducted by using acid-base titration method. Five milliliters of barium hydroxide solution (0.1 N) were added into 0.1 g of CO_2–α-CD inclusion complex in a 30-ml Erlenmeyer flask filled previously for the release experiment. The mixture was shaken gently to fully dissolve the inclusion complex and titrated drop-wise to a phenolphthalein endpoint with hydrogen chloride solution (0.28 N) using a 2-ml micrometer syringe (S-1200, Gilmont Instruments Inc., Illinois, USA). The inclusion ratio is defined as the molar ratio of the included CO_2 to α-CD (mol CO_2/mol α-CD).

IR spectra of the complex powder with Fourier transform infrared spectroscopy (FT-IR)

A Shimadzu FTIR-8300 Spectrophotometer (Shimadzu Corp., Kyoto, Japan) equipped with HYPER-IR software was used for recording the IR spectra of the α-CD

and inclusion complexes. The KBr pellet method was employed and measurement was performed at room temperature. The KBr pellets of samples were prepared with a hydrostatic pressure of 6–7 ton cm^{-2} for 1 min. The scanning range was set at 400–4000 cm^{-1} and 20 scans were collected with a spectral resolution of 4.0 cm^{-1}.

Powder X-ray diffractometry (PXRD)

Powder X-ray diffraction profiles were recorded on a Shimadzu LabX XRD 6000 X-ray diffractometer (Shimadzu Corp., Kyoto, Japan). Measurements were performed using Ni-filtered CuKα radiation ($\lambda = 1.5418$ Å) operating at 30 kV – 35 mA. Samples were scanned at a scanning speed of 2°/min over a diffraction angular range of $3° < 2\theta < 30°$. The divergence slit, receiving slit and time constant were set at 1°, 0.3 mm and 1 s, respectively.

Scanning electron microscopy (SEM)

Microscopy of α-CD and the inclusion complexes of CO$_2$ and α-CD was performed using a JEOL JSM-6060 scanning electron microscope (JEOL Ltd., Tokyo, Japan). The samples were deposited on double-coated carbon conductive tape adhered to the specimen mounts and analyzed at 2.0 kV with a spot size of 30.

Results and discussion

Formation of the inclusion complexes at different initial moisture contents and CO$_2$ pressures

Figure 1 represents the encapsulation time-courses of CO$_2$ in solid state α-CDs. In the beginning of encapsulation reaction, inclusion ratio, N, increased linearly with the time, and gradually approached an equilibrium value which was dependent on the pressure of CO$_2$. The main aspect of interest was the effect of initial moisture content of α-CD on the encapsulation behavior.

Generally, increase of initial moisture content had noticeably raised the encapsulation rate. The encapsulation time-courses were correlated by the following first order encapsulation equation:

$$N = N_{\max}(1 - \exp[-(k_e \cdot t)]) \tag{1}$$

where N (mol CO$_2$/mol α-CD) is the inclusion ratio at time t (s), k_e (s^{-1}) the inclusion rate constant and $N_{\max}$ (mol CO$_2$/mol α-CD) the maximum inclusion ratio. Those parameters calculated by Equation (1) are listed in Table 1. The increase of initial moisture content appreciably increased the values of k_e. Encapsulation of CO$_2$ into α-CD with 2 wt% moisture was found occurring at the lowest rate. Increase of initial moisture content to 10 wt% has speeded up the encapsulation rate drastically. This might be attributable to the relative polarity between water molecules and CO$_2$ which enhances the driving force for encapsulation. It is known that α-CD forms a hexahydrate with four water molecules located outside and two included in the cavity [13–15]. These two included water molecules were referred to as "high-energy water" and are energetically not comparable with water molecules in the bulk phase [16, 17]. Thus the release of these water molecules from the cavity should result in favorable enthalpic and entropic contributions to the Gibbs free energy, giving an important effect on the inclusion driving force [17–19].

In spite of that, further increment to 30 wt% moisture did not continue to escalate the encapsulation rate to a higher value. The effect of initial moisture content on $N_{\max}$ value followed a similar trend as its effect on the encapsulation rate. No convincing reasoning could be achieved for explaining this trend.

Further, encapsulation of CO$_2$ into solid α-CD was compared between two encapsulation pressures of 1 and 3 MPa CO$_2$. For every initial moisture content studied, higher CO$_2$ pressure was observed to have constantly accelerated the encapsulation rate and heightened the maximum inclusion ratio. The pressure dependence of the intensity of IR vibrational band of CO$_2$ incorporated
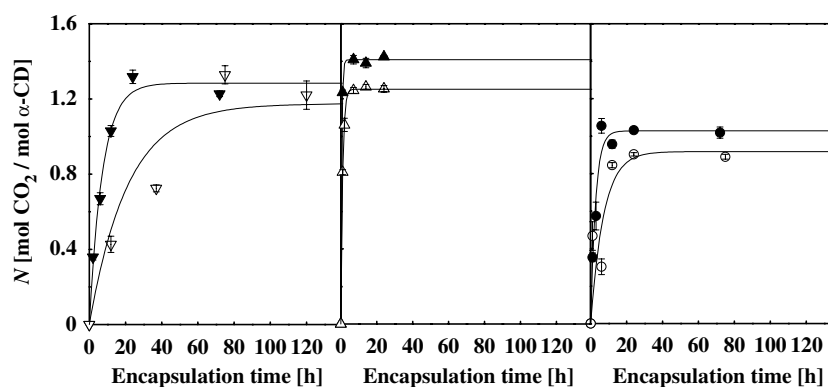

Figure 1. Encapsulation behavior of CO$_2$ into α-CD with different initial moisture contents at 25 °C. Initial moisture content of α-CD: ▼▽, 2 wt%; ▲△, 10 wt%; ●○, 30 wt%. Closed symbols represent encapsulation carried out under pressure of 3 MPa CO$_2$ and open symbols under 1 MPa CO$_2$.

Table 1. Parameters of encapsulation time-courses of inclusion complexes prepared from solid α-CD of various initial moisture contents at 25 °C

Initial moisture content of α-CD	2 wt% (vacuum dried)		10 wt%		30 wt%	
Carbon dioxide pressure [MPa]	1	3	1	3	1	3
N_{max} [mol CO_2/mol α-CD]	1.17	1.28	1.25	1.41	0.92	1.03
k_e [s^{-1}]	170	490	3600	7500	500	1300

in *p*-t-butylcalix[4]arene cavity was revealed by Graham *et al.* [20] which also implied the pressure dependence of the inclusion ratio of the particular complex.

In saturated α-CD solution, the amount of α-CD collected in the form of precipitate of complex crystals was found increasing linearly with time in the initial stage of inclusion period and then asymptotically approaching an equilibrium value as shown in Figure 2. Figure 2 is a plot of the percentage of α-CD precipitated in the form of inclusion complex crystal versus the encapsulation time. The percentage of α-CD precipitated was the mass ratio of α-CD in the complex crystal to the initial α-CD dissolved in the saturated solution. The inclusion ratios of the precipitated crystals stayed at around 0.97–1.08 mol CO_2/mol α-CD. It is conceivable that α-CD in aqueous phase crystallized and precipitated only when its inclusion complex with CO_2 was formed, because this inclusion complex had lower water solubility compared to the α-CD molecule alone.

Release of CO_2 from the complex powder

The difference in initial moisture content of α-CD during encapsulation resulted in variation in moisture content of inclusion complexes. Hence, except for inclusion complex prepared from α-CD with 2 and 10 wt% initial moisture, alignment of moisture content to the range of 7–11 wt% was carried out by vacuum drying at room temperature (20 ± 5 °C) for 2 h before subjection to release experiment. Quantification of CO_2 prior to vac-

uum drying procedure has confirmed that the drying step had negligible effect on the inclusion ratio. The moisture contents of inclusion complexes from α-CD with 2 and 10 wt% initial moisture remained within the range of 1–3 and 7–11 wt%, respectively.

The inclusion complexes were subjected to a 1-week length release experiment at 25 °C and at relative humidities of 5, 33 and 75%, respectively. The inclusion complex powders encapsulated for 72 h were used, as almost all the inclusion complexes were found to have reached the equilibrium by this encapsulation time. Table 2 shows the initial inclusion ratios and the standard deviations of the inclusion complex powders used for release experiment.

Figure 3 shows the release time-courses of CO_2 from the inclusion complex powders. The characteristics of the CO_2 release are dependent not only on the relative humidity of the storage atmosphere but also the initial moisture content of α-CD during encapsulation. To estimate the release rate of CO_2, the following Avrami's equation was applied for correlation of the release time-courses:

$$R = \exp(-[k_r \cdot t]^n) \quad (2)$$

where R (−) is the retention of CO_2 in the complex powder at time t (s), k_r (s^{-1}) the release rate constant and n (−) the release mechanism parameter. The correlations of release time-courses are presented in Figure 3 by the solid lines, indicating well correlations of the release characteristics of CO_2 at various conditions. This particular equation was recently reported of its applications to the release time-courses of different encapsulated compounds prepared by spray drying [21, 22]. The n value in the Avrami's equation is the parameter representing the release mechanism that describes the shape of a release time-course. Theoretically, the parameter
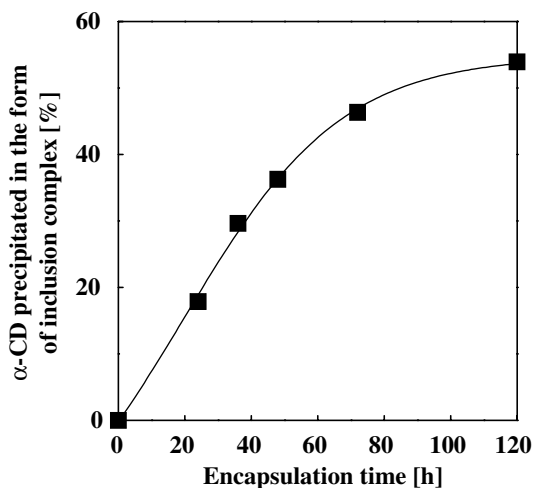

Figure 2. Precipitation of complex crystal from saturated α-CD solution. Encapsulation is performed under 3 MPa CO_2 at 25 °C.

Table 2. Initial inclusion ratios of various inclusion complexes prepared for 72 hours at 25 °C

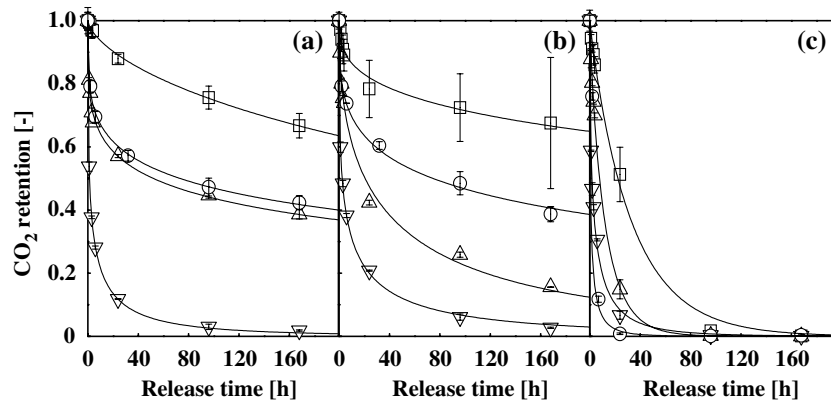| Initial moisture content of α-CD during encapsulation	Initial inclusion ratio [mol CO_2/mol α-CD]	Standard deviation
2 wt%	1.12	0.07
10 wt%	1.31	0.07
30 wt%	1.08	0.09
Saturated solution (87.3 wt%)	1.06	0.02

Figure 3. The release time-courses of CO_2-α-CD inclusion complexes at 25 °C and different storage humidities. (a) 5% RH, (b) 33% RH, (c) 75% RH. Initial moisture content of α-CD during encapsulation reaction: □, saturated α-CD solution; ○, 30 wt%; △, 10 wt%; ▽, 2 wt% (vacuum dried).

$n = 0.54$ corresponds to diffusive release and $n = 1$ to a first order release kinetics which generally occurs when the guest compound is actually a solution. By a linear plot of $\ln(-\ln R)$ versus $\ln t$ from Equation (2), we obtained the n value from the inclination and the release rate constant, k_r from the interception at $\ln t = 0$. The relations of the release rate constant, k_r and release mechanism parameter, n are summarized against the storage humidity in Figures 4 and 5.

Effect of initial moisture content of α-CD during encapsulation on release characteristic

Generally, stability of CO_2–α-CD inclusion complexes showed vivid dependency on initial moisture content of α-CD during encapsulation. At storage humidity of 5% RH, the retention of CO_2 (stability) of inclusion complexes against release time increased with increase of the initial moisture content. The most stable time-course of CO_2 retention was shown by inclusion complex pre-

pared from saturated α-CD solution with the lowest release rate constant of 5.33 s^{-1}. The CO_2 retention decreased only to 0.67 over the release for 1 week (168 h). Meanwhile, the inclusion complex from α-CD with 2 wt% moisture displayed the lowest stability with almost complete release at the end of the duration. The k_r of this particular inclusion complex gave a value as high as up to 1089 s^{-1}, which was three orders greater than that of the inclusion complex prepared from saturated α-CD solution. α-CD containing 30 wt% initial moisture produced inclusion complex more stable than that from α-CD with 10 wt% initial moisture. Nevertheless the difference was trivial whereby the release rate constants differed by only a small magnitude between one another.

When stored at humidity up to 33% RH, the release time-courses of these inclusion complexes varied more clearly with the initial moisture content. The decline in stability of these inclusion complexes still followed the
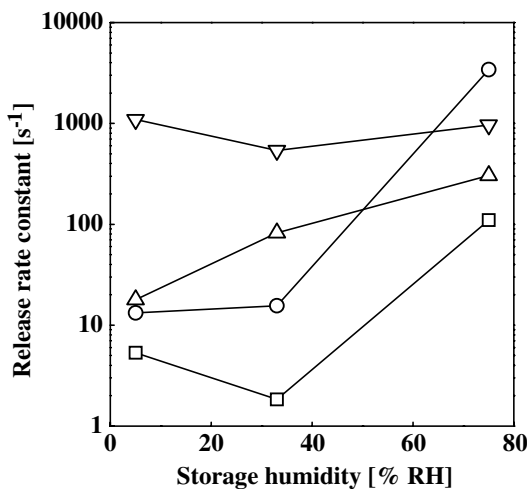

Figure 4. Release rate constants (k_r) calculated from the Avrami's equation for CO_2–α-CD complexes stored at 25 °C and various relative humidities. Inclusion complex prepared from: □, saturated α-CD solution and α-CD with initial moisture content of ○, 30 wt%; △, 10 wt%; ▽, 2 wt% (vacuum dried).
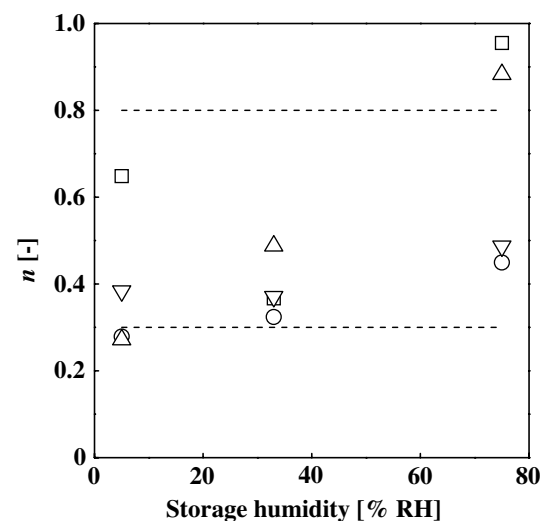

Figure 5. Release mechanism factors (n) calculated from the Avrami's equation for CO_2–α-CD complexes stored at 25 °C and various relative humidities. Inclusion complex prepared from: □, saturated α-CD solution and α-CD with initial moisture content of ○, 30 wt%; △, 10 wt%; ▽, 2 wt% (vacuum dried).

similar order as shown in storage humidity of 5% RH. The CO_2 retention given by the inclusion complex prepared from saturated α-CD solution has distinctly huge standard deviation which is expanding towards the end of storage period.

At 75% RH, generally increase was observed in the release rate constants. Taking the experimental error into consideration, except for the inclusion complex from saturated α-CD solution, all the inclusion complexes were considered to have released CO_2 at similar rate. Almost complete release of CO_2 was observed in all the inclusion complexes after 96 hours of storage.

Effect of storage humidity on release characteristic

In general, effect of storage humidity on the release characteristics of CO_2–α-CD complexes is rather conclusive. The dependence of release on storage humidity could be considered simple as release of CO_2 increased with increasing storage humidity. At low storage humidity of 5% RH, inclusion complexes were generally far more resistant to release, especially for the complex powders prepared at higher initial moisture content, as shown in Figure 4. As the storage humidity rose higher, the inclusion complex became less stable and the release rate of CO_2 became higher. Considering the inclusion complexes prepared from saturated α-CD solution and α-CD with 30 wt% initial moisture, the acceleration of release rate was not observed from 5% RH to 33% RH of storage humidity. Nevertheless, increase of release rate constant, k_r occurred by two orders of magnitude when these inclusion complexes were kept at 75% RH, as shown in Figure 4. As for inclusion complex from α-CD with 10 wt% initial moisture, release rate heightened moderately in response to the rising storage humidity from 5 to 75% RH. For inclusion complex from α-CD with 2 wt% initial moisture, the release rates were almost independent of storage humidities between 5 and 75% RH. Generally, k_r values of inclusion complex prepared at lower initial moisture content are higher, indicating the weak host–guest interaction between CO_2 and α-CD. Since most of the release mechanism factors, n fell within the range of 0.30–0.80, as shown in Figure 5, it indicated that the release mechanism of CO_2 from these inclusion complexes may be approximated to diffusion. In other words, the diffusion seemed to have been promoted by moisture in storage environment. Similar effect of relative humidity on release of *d*-limonene incorporated in modified CD bonded to cellulose has been reported by Rehmann *et al.* [19].

Fourier Transform Infrared Spectroscopy (FT-IR)

FT-IR was utilized in determining the state of existence of CO_2 after encapsulation. CO_2 is known to tend to react with alcohol to form carbonic acid. Hence, during encapsulation in the presence of water, CO_2 has been suspected to have chemically bound to the primary hydroxyl groups of α-CD rather than being encapsulated into the α-CD cavity. If this would be true, the inclusion complexes prepared might be improper to be claimed as "inclusion complex". The FT-IR confirmed the complex formation and as well the host:guest interaction between CO_2 and α-CD. Infrared spectra of α-CD and CO_2–α-CD complex prepared from saturated α-CD solution are presented in Figure 6 to serve reference purpose.

The carbon–oxygen double bond, $C=O$ in organic compounds such as esters, aliphatic and aromatic aldehydes, etc, was found within the range of wavenumber from 1670 to 1810 cm^{-1} [23]. Its position varies slightly depending on what sort of compound it is in. Therefore, if CO_2 were to chemically bind to the primary hydroxyl groups of α-CD, absorption band would be observed at wavenumber from 1670 to 1810 cm^{-1}. From Figure 6, absorption band was found at wavenumber slightly lower than this range ($\approx$1640 cm^{-1}). However, since similar one was also being observed in IR spectrum of α-CD without CO_2 encapsulation, these particular absorption bands were concluded not being encapsulated CO_2.

The linear CO_2 molecule has no net dipole moment at rest. Nonetheless, it has stretching (symmetric: $v_1 = 1388$ cm^{-1}, asymmetric: $v_3 = 2349$ cm^{-1}) as well as non-linear vibrational modes (bending: $v_2 = 667$ cm^{-1}) that produce a variable dipole moment and allow the absorption of infrared light [24]. The symmetric stretching vibration at around 1388 cm^{-1} is infrared inactive which can only be detected using Raman spectroscopy [25]. Comparison of IR spectra was made between α-CD and inclusion complexes focusing on wavenumber around 2349 cm^{-1}. All the α-CDs treated with encapsulation contained a strong vibrational band at 2334–2338 cm^{-1} that was not observable in the spectrum of uncomplexed α-CD. The wavenumber at
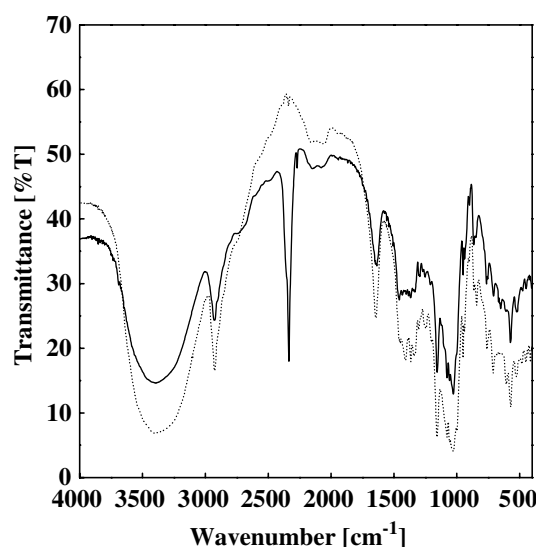

Figure 6. Infrared spectra (4000–400 cm^{-1}) of ordinary α-CD and inclusion complex prepared from saturated α-CD solution. Dashed line: uncomplexed α-CD; solid line: inclusion complex from saturated α-CD solution.

which this particular absorption band was caused was red-shifted by 11–15 cm^{-1} from that of the pure gaseous CO_2. Graham et al. [20] reported that the vibrational spectrum of CO_2 resides in the p-t-butylcalix[4]arene cavity forming a host:guest inclusion complex was red-shifted. This phenomenon is also similar to the vibrational spectrum of gaseous CO_2 trapped in a KBr pellet described by Keresztury et al. [26]. The existence of CO_2 as gaseous guest compound in α-CD was conclusive as the symbolic band of gaseous CO_2 at ≈2338 cm^{-1} was only observable in IR spectra of inclusion complexes. The pressurized condition of CO_2 and the inclusion of CO_2 inside the α-CD cavity may partly be responsible for the red-shift of CO_2 absorption band.

A strong v_3CO_2 band would always be accompanied by a sharp, intense bending band at 667 cm^{-1}. Pressing of CO_2 into KBr pellet showed great reduce in intensity of this band [26]. In the IR spectra of inclusion complexes, the bending vibration (deformation vibration) of CO_2 at this particular wavenumber was difficult to distinguish as it falls within the fingerprint region (500–1500 cm^{-1}) which consists of absorptions due chiefly to all kinds of bending vibration within all the molecules in the samples. The pattern of spectrum in this region was in fact similar in all the IR spectra. Most probably the absorption band caused by CO_2 bending vibration has undergone some shrinkage and thus been overlapped. The possible causes of the suppression of this band were conceived of to be the pressurization of inclusion complexes into the KBr pellet and the encapsulation of CO_2 within the α-CD cavity which may both have hindered the bending vibration of CO_2.

Powder X-ray diffractometry (PXRD)

Figure 7 compares the powder X-ray diffraction patterns of uncomplexed α-CDs (a and b) and inclusion complexes of different initial moisture contents (c, d, e and f). Sharp peaks in the diffractograms suggested that the α-CD and as well the inclusion complexes were present as crystalline materials. Comparison between uncomplexed α-CDs and inclusion complexes revealed that inclusion of CO_2 into the α-CD cavity changed the crystal lattice structure of α-CD and diffraction peaks were distinguished more clearly. More importantly, a new characteristic peak at diffraction angle around 13.3° appeared only in the inclusion complexes, as shown in Figures 7(c) to (f). Although PXRD patterns of uncomplexed α-CDs of different moisture content differed slightly from each other as reported by Nakai et al. [14], the characteristic peak at 13.3° could not be found. This implied that the peak at 13.3° was resulted from the inclusion of CO_2 in α-CD but not from hydration of α-CD. Nevertheless, since the overall changes in PXRD patterns were rather modest, the CO_2 molecule was presumably too small to induce great changes in the crystal lattice structure of α-CD. Considering only the inclusion complexes, the diffractograms were to some

extent similar to each other regardless of the moisture contents.

Scanning Electron Microscopy (SEM)

Figure 8 shows the SEM microphotographs of α-CD and various inclusion complexes. Comparing inclusion complexes prepared at initial moisture of 2 and 10 wt%, the added moisture might have caused slight agglomeration of inclusion complex which could be responsible for improvement of complex stability against release. As for inclusion complex prepared at initial moisture of 30 wt%, stability enhancement may be accredited to the physical aspect. Since the release mechanism was mostly diffusion limited (from the n values calculated earlier), agglomeration of crystals forming huge lumps might have further retarded the diffusion of CO_2 to the outer environment. In saturated solution of α-CD, inclusion complex crystallized and grew slowly inside water. The
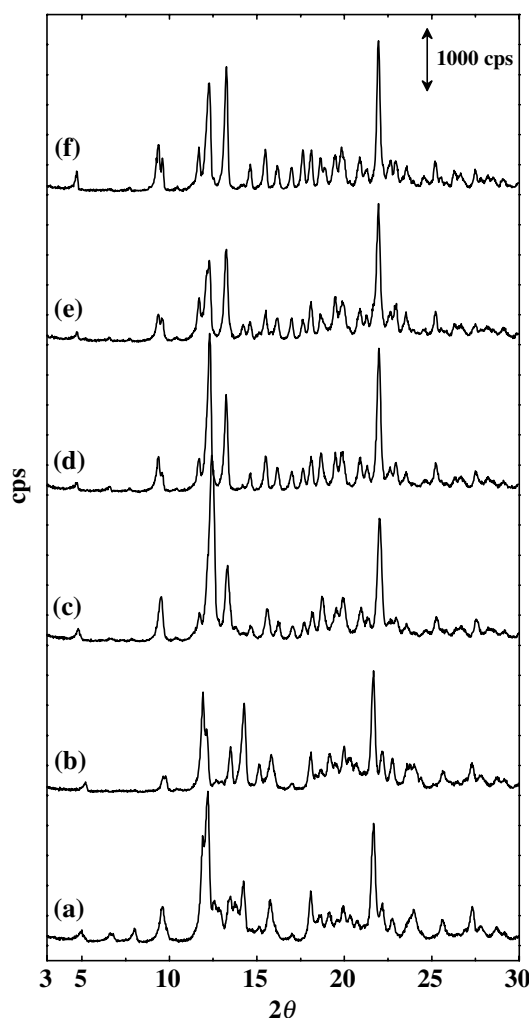

Figure 7. Powder X-ray diffraction patterns of α-CDs and inclusion complexes. α-CD with (a), 2 wt% moisture; (b), 10 wt% moisture, and inclusion complexes prepared from α-CD with various initial moisture contents: (c), 2 wt%; (d), 10 wt%; (e), 30 wt% and (f), saturated α-CD solution. When measurements are being carried out, samples (a) and (c) contain 1–3 wt% moisture and the rest contain 7–11 wt% moisture.
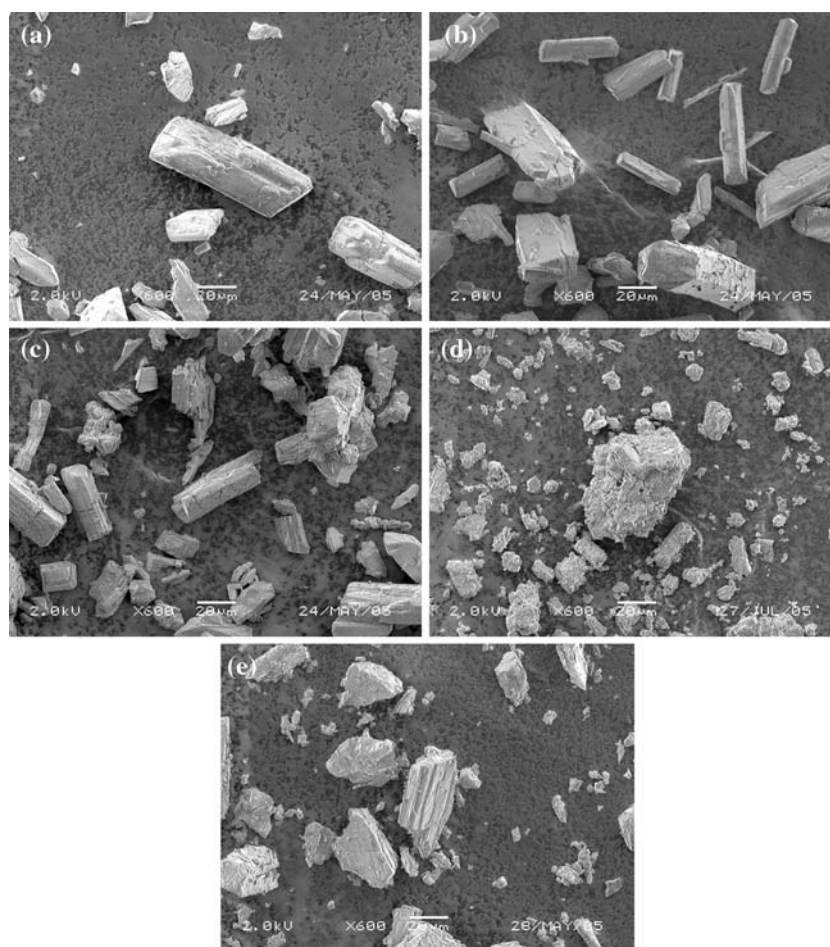
Figure 8. SEM photographs of (a), α-CD and inclusion complexes prepared from α-CD with different initial moisture contents: (b), 2 wt%; (c), 10 wt%; (d), 30 wt% and (e), saturated α-CD solution.

crystals grew layer by layer to form a systematic structure which was thought to have effectively decelerated the release rate.

Conclusion

In conclusion, encapsulation was affected by initial moisture content of α-CD and the CO_2 pressure. Inclusion complex stability was dependent on initial moisture content of α-CD during encapsulation and storage humidity. CO_2 was proved to be encapsulated within α-CD cavity and existed in gaseous state. CO_2-α-CD inclusion complex was a true inclusion complex that differed in crystal lattice structure from the uncomplexed α-CD. Inclusion complex stability was postulated to be related to the physical structure of the inclusion complex itself.

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