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Peracylated β-Cyclodextrins as Novel Sustained-release Carriers for a Water-soluble Drug, Molsidomine

KANETO UEKAMA, TAKASHI HORIKAWA, MASAYUKI YAMANAKA AND FUMITOSHI HIRAYAMA

Faculty of Pharmaceutical Sciences, Kumamoto University, 5-1 Oe-honmachi, Kumamoto 862, Japan

Abstract—Peracylated β-cyclodextrins with different alkyl chains (acetyl-octanoyl) were prepared by acylating all hydroxyl groups of β-cyclodextrin (β-CyD), and their physical properties were evaluated. These hydrophobic β-CyDs decreased the release rate of molsidomine, a peripheral vasodilator, in proportion to the lengthening of alkyl chain and suppressed a peak plasma level of molsidomine following oral administration of peracylated β-CyD complexes to dogs. Among the peracylated β-CyDs tested, perbutanoyl-β-CyD maintained sufficient plasma drug levels for a long period of time, while other peracylated β-CyDs having shorter or longer chains were inappropriate to control the in-vivo release behaviour of molsidomine. The prominent retarding effect of perbutanoyl-β-CyD was ascribable to the appropriate mucoadhesive property and hydrophobicity, compared with other peracylated β-CyDs. The present results suggest that perbutanoyl-β-CyD is particularly useful in modifying the release rate of water-soluble drugs as a novel slow-release carrier.

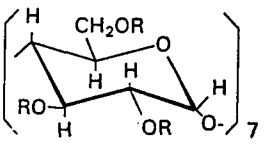
There has been increasing interest in optimizing the efficacy of drug activity through the use of rationally designed drug carrier materials. Cyclodextrin (CyD) is a potent candidate for such a role, modifying physical, chemical and biological properties of drug molecules through the formation of inclusion complexes (Uekama & Otagiri 1987; Szejtli 1988). Recently, various kinds of CyD derivatives have been prepared to extend the physicochemical properties and inclusion capacities of natural CyDs as multi-functional drug carriers (Duchêne 1991; Uekama et al 1991; Hirayama 1993). For example, hydrophilic CyD derivatives such as hydroxyalkylated β-CyDs have been used for improvements of low solubility, dissolution rate and bioavailability of poorly water-soluble drugs (Müller & Brauns 1985; Pitha & Pitha 1985; Yoshida et al 1990). On the other hand, hydrophobic CyD derivatives such as ethylated β-CyDs are reported to be useful as slow-release carriers for water-soluble drugs such as diltiazem hydrochloride (Horiuchi et al 1990), isosorbide dinitrate (Hirayama et al 1988) and buserelin acetate (Uekama et al 1989). In this study, we prepared a new series of hydrophobic peracylated CyD derivatives, heptakis(2,3,6-tri-*O*-acyl)-β-CyDs, with different chain lengths (acetyl-octanoyl), and evaluated their pharmaceutical functions. Attention was given to highly hydrophobic and mucoadhesive properties of peracylated β-CyDs, anticipating more effective slow-release carriers for oral preparations of water-soluble drugs. Molsidomine, a peripheral vasodilator which is soluble in water (0.25 g dL^{-1} at 25°C) and has a short biological half-life (2.1–2.7 h) in man (Yashiki et al 1985) was used as a model drug.

Materials and Methods

β-CyD and molsidomine were donated by Nihon Syokuhin Kako Co. Ltd (Tokyo, Japan) and Takeda Chemical Ind. Ltd (Osaka, Japan), respectively. Peracylated β-CyDs with different alkyl chains (acetyl-octanoyl) were prepared by acylating all hydroxyl groups of β-CyD, using corresponding acid anhydrides in pyridine solution. A single component, heptakis(2,3,6-tri-*O*-acyl)-β-CyD, was isolated by silica gel column chromatography, and characterized by nuclear magnetic resonance and fast-atom bombardment mass spectrometry, and elemental analysis. The detailed preparation and characterization of peracylated β-CyDs will be reported elsewhere. The complexes of molsidomine with peracylated β-CyDs in a 1 : 1 molar ratio were prepared by the kneading method using ethanol as a solvent, and characterized by differential scanning calorimetry (DSC) and powder X-ray diffractometry, in the same manner as reported previously (Horiuchi et al 1990). The release rate of molsidomine from gelatin capsules (volume 1.0 cm^3) was measured according to the paddle method of the dissolution test in the Japanese Pharmacopoeia XII (JP XII) in 2nd fluid (pH 6.8, 500 mL) at a stirring speed of 100 rev min^{-1}. In the in-vivo absorption studies, the gelatin capsules containing the complexes (equivalent to 10 mg molsidomine) were orally administered to four beagle dogs (male, 10–14 kg) that were fasted for more than 24 h. High-performance liquid chromatography (HPLC) was employed for the determination of molsidomine in dissolution medium and dog plasma, according to the method described by Mitani et al (1985). The HPLC conditions were as follows: a Hitachi 655A-11 chromatograph (Tokyo, Japan); YMC AQ-312 ODS column (Kyoto, Japan); mobile phase, 0.01 M sodium acetate buffer/acetonitrile (4 : 1 v/v); detection, 313 nm; flow rate, 1.0 mL min^{-1}. The adhesive properties of peracylated β-CyDs were evaluated by means of the force of detachment; samples were dissolved in acetone and dried for different periods of time under atmospheric pressure to make gels with different concentrations of the substrates, and a probe having a plane surface (area: 0.2 cm^2) was allowed to stand in contact with the surface of the gels. The force required to detach the probe from the gel surface

Correspondence: K. Uekama, Faculty of Pharmaceutical Sciences, Kumamoto University, 5-1 Oe-honmachi, Kumamoto 862, Japan.

Table 1. Some physical properties of peracylated β-CyDs at 25°C.



β-CyDs	R	Melting point (°C)	$[\alpha]_D$[a]	Solubility[c] (mg dL^{-1})
β-CyD	H	280	163[b]	119.0
Peracetyl-β-CyD	$COCH_3$	201–202	125	823.0
Perpropanoyl-β-CyD	COC_2H_5	168–169	106	423.5
Perbutanoyl-β-CyD	COC_3H_7	126–127	100	219.8
Perpentanoyl-β-CyD	COC_4H_9	54–56	91	283.0
Perhexanoyl-β-CyD	COC_5H_{11}	(oil)	82	3.7
Peroctanoyl-β-CyD	COC_7H_{15}	(oil)	73	[d]

[a] In chloroform. [b] In water. [c] In 80% (v/v) ethanol/water. [d] Could not be determined because of the low solubility.

was measured by a probe-tack tester (Rigaku Kogyo Co. Tokyo, Japan) under the following conditions: contact time, 10 s; shear rate, 5 mm s^{-1}; amount loaded on the probe, 45 g cm^{-2}. The viscosity of peracylated β-CyDs in acetone was measured using an Ostwald viscometer at 20°C.

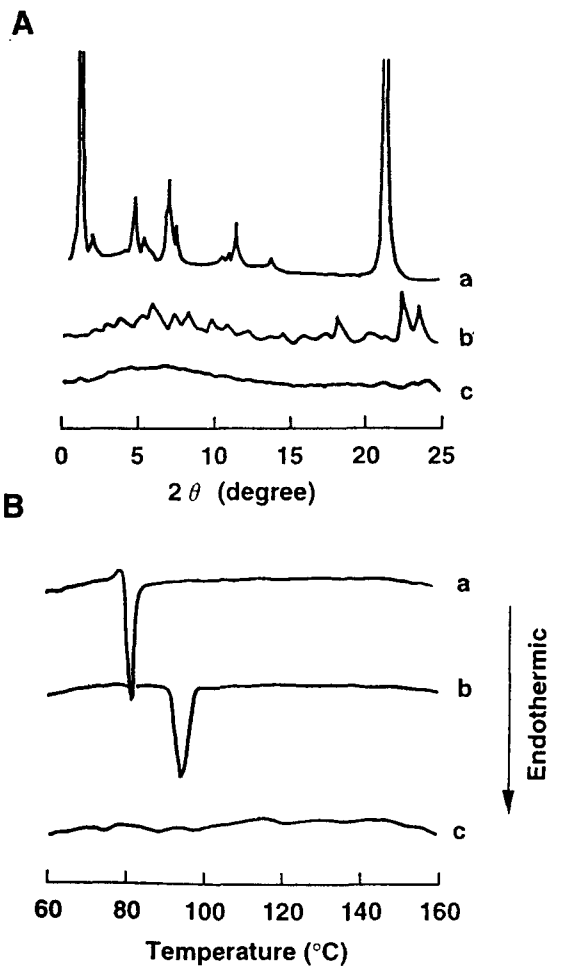


FIG. 1. Powder X-ray diffraction patterns (A) and DSC thermograms (B) of molsidomine-perbutanoyl-β-CyD complex system. a. Molsidomine, b. perbutanoyl-β-CyD, c. complex of molsidomine with perbutanoyl-β-CyD.

Results and Discussion

Table 1 summarizes and compares some physical properties of peracylated β-CyDs with those of the parent β-CyD. Because of their poor aqueous solubility, the solution properties of peracylated β-CyDs were measured after dissolving them in appropriate organic solvents. With increasing alkyl chains, melting point, $[\alpha]_D$, and solubility decreased. The concentrated solutions of peracylated β-CyDs in solvents such as ethanol, acetone and chloroform were highly viscous and sticky, and gelation took place upon evaporation of the solvents. Since these properties were thought to be particularly useful in a slow-release drug carrier, the solid complexes of peracylated β-CyDs with water-soluble molsidomine were prepared and their in-vitro and in-vivo release behaviours were compared.

Fig. 1 shows a typical example of powder X-ray diffraction patterns and DSC thermograms of molsidomine-perbutanoyl-β-CyD complex system. The diffraction pattern of the complex was apparently different from that of each component, and showed a halo pattern. Similarly, the DSC thermogram of the complex showed no endothermic

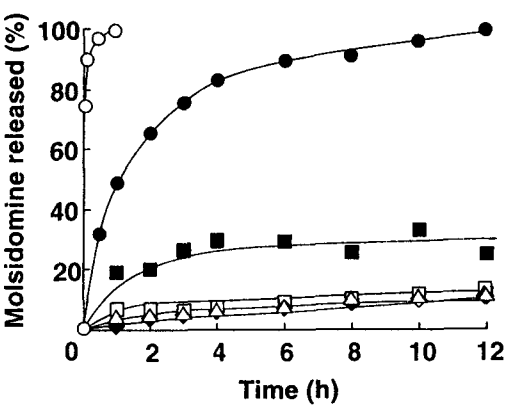


FIG. 2. Release profiles of molsidomine from capsules containing molsidomine or its peracylated β-CyD complexes (equivalent to 10 mg molsidomine) in JP XII 2nd fluid (pH 6.8) at 37°C. ○ Molsidomine alone (starch diluent), ● peracetyl-β-CyD complex, ■ perpropanoyl-β-CyD complex, □ perbutanoyl-β-CyD complex, △ perpentanoyl-β-CyD complex, ◆ perhexanoyl-β-CyD complex.

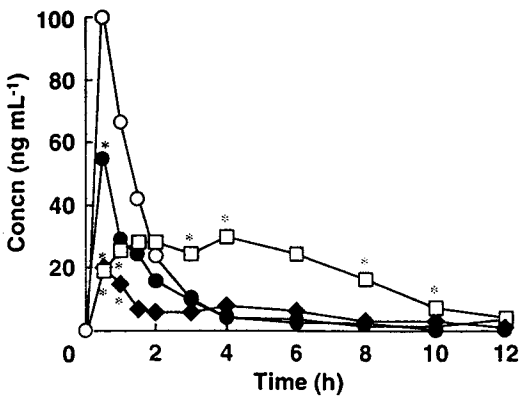


FIG. 3. Plasma levels of molsidomine after oral administration of capsules containing molsidomine or its peracylated β-CyD complexes (equivalent to 10 mg molsidomine) in dogs. ○ Molsidomine alone (starch diluent), ● peracetyl-β-CyD complex, □ perbutanoyl-β-CyD complex, ◆ perhexanoyl-β-CyD complex. Each value represents the mean of 3–6 dogs. $*P < 0.05$ compared with molsidomine alone.

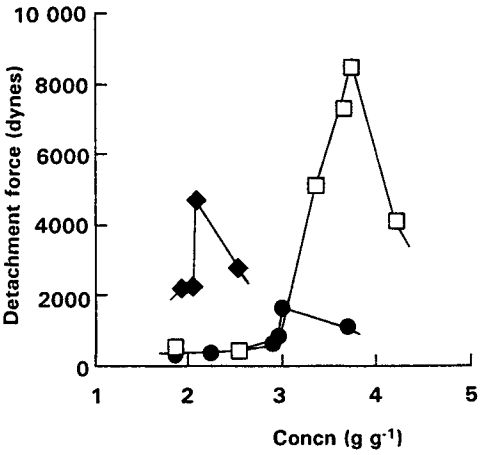


FIG. 4. Force of detachment of peracylated β-CyDs as a function of peracylated β-CyD concentrations in acetone, measured by a probe-tack tester at 25°C. ● Peracetyl-β-CyD, □ perbutanoyl-β-CyD, ◆ perhexanoyl-β-CyD.

peaks due to the melting of both components, suggesting that an amorphous complex was formed in the solid state. Fig. 2 shows the release profiles of molsidomine from capsules containing the drug and starch as a diluent or its peracylated β-CyD complexes into the JP XII 2nd fluid (pH 6.8). The release rate from the starch mixture was very fast, due to the high aqueous solubility of the drug. On the other hand, the drug release was markedly retarded by the complexation with peracylated β-CyD complexes in decreasing order of solubility of acylated β-CyDs. Since the peracylated derivatives, which are longer than the hexanoyl moieties, hardly released molsidomine under the experimental conditions, peracetyl-, perbutanoyl- and perhexanoyl-β-CyDs were used in the subsequent in-vivo absorption studies. Fig. 3 shows the plasma concentration of molsidomine vs time curves obtained after oral administration of a single dose of capsules containing either molsidomine or its peracylated β-CyD complexes (equivalent to 10 mg molsidomine) to dogs. Table 2 summarizes the pharmacokinetic parameters obtained from the data of Fig. 3, where the mean residence times (MRT) in the systemic circulation calculated by moment analysis (Yamaoka et al 1981) are also listed. As shown in Fig. 3, the absorption of molsidomine from the capsules was rapid, with a short biological half-life in the plasma, while sustained-release patterns were evident for the absorption of molsidomine from the peracylated β-CyD complexes. This is probably due to the slow rate of drug release. It is noteworthy that only the perbutanoyl-β-CyD complex produced an increase in area under the plasma concentration curve (AUC), up to 12 h post-administration, and significantly prolonged the time (t_{max}) required to reach the maximum plasma level (C_{max}), compared with molsidomine alone. In addition, the MRT value of the perbutanoyl-β-CyD complex was about twice that of molsidomine alone, indicating a possible sustained-release preparation in the dog model. In the case of peracetyl-β-CyD complex, however, the plasma levels were maintained at a comparatively low concentration, in spite of the lower hydrophobicity of the peracylated β-CyDs employed. This indicates that not only hydrophobicity but also additional properties of carrier material may be responsible for the control of in-vivo release of molsidomine.

Since the prominent retarding effect of perbutanoyl-β-CyD with increase in AUC appeared to be due to its longer residence time in the gastrointestinal tract, factors responsible for this mechanism were examined. As a measure of mucoadhesion, the force of detachment of peracylated β-CyDs was evaluated. Fig. 4 shows the detaching force of CyD gels as a function of its amount in acetone. The detaching force of CyDs increased with increase in its concentration and then decreased after the peaks. The decrease of the force at higher CyD concentrations can be ascribed to the deposition of CyD crystals or oils on the surface of gels. It is apparent that the maximum adhesional force of perbutanoyl-β-CyD was considerably higher than those of other peracylated β-CyDs. The viscosity

Table 2. Pharmacokinetic parameters of molsidomine after oral administration of capsules containing molsidomine or its acylated β-CyD complexes (equivalent to 10 mg molsidomine) in dogs.

System	C_{max} (ng mL^{-1})	t_{max} (h)	AUC (h ng mL^{-1})	MRT (h)
Molsidomine	102.1 ± 15.4	0.6 ± 0.2	153.5 ± 44.9	1.8 ± 0.4
Peracetyl-β-CyD	54.7 ± 4.5*	0.5 ± 0.1	90.7 ± 17.3	1.9 ± 0.5
Perbutanoyl-β-CyD	42.0 ± 7.8*	3.3 ± 1.0*	225.4 ± 66.2	4.5 ± 0.4*
Perhexanoyl-β-CyD	30.3 ± 14.9*	4.8 ± 2.8	67.2 ± 6.3	4.5 ± 1.9

Each parameter is expressed as the mean±s.e. of 3–6 dogs. $*P < 0.05$ compared with values for molsidomine.

of peracylated β-CyDs increased with increasing alkyl chains; peracetyl-β-CyD (1.7 cP) < perbutanoyl-β-CyD (2.7 cP) < perhexanoyl-β-CyD (3.6 cP) at a concentration of 0.2 M in acetone. These results suggest that the retarding effect of perbutanoyl-β-CyD can be ascribed to the appropriate mucoadhesive property and hydrophobicity.

As mentioned above, perbutanoyl-β-CyD sufficiently maintained the plasma drug levels for a long period of time, while other acylated β-CyDs having shorter or longer chains were inappropriate for practical application, because of low absorption. It is also noteworthy that the perbutanoyl-β-CyD complex system showed no appreciable change in AUC value even in non-fasted dogs (data were not shown). Nevertheless, the present results clearly suggest that peracylated β-CyDs may be useful to modify the release rate of various water-soluble drugs as a multi-functional carrier.

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