CHAPTER I

INTRODUCTION

Rationale

Cyclodextrins (CDS) are valuable material and well known for their ability to encapsulate various molecules, conferring on them new physicochemical properties. However, the greatest interest in cyclodextrins concern with increasing in water solubility, stability and bioavailability.

Indomethacin (IDM) is slightly soluble in water, (0.4 mg/ml) (Brien, MaCauley and Cohen, 1984) was chosen as a model drug. This drug exists in several polymorphs, therefore, possess different in physicochemical properties including solubility. Moreover, the dissolution criteria of IDM capsule according to USP XXII requirement is also high, (80% of IDM released in 20 mins.) thus the rapid solubility should be achieved. The rapid dissolution rate of the drug from the dosage form is, therefore, considerably important in manufacture of this capsule product. It was claimed that the products which produced high drug concentration at the site of absorption could exhibit higher bioavailability, thus reduce in gastric irritation and in dose administered.

An effort had been made to increase the dissolution rate of IDM with CDS (Hamada et al., 1975; Kurozumi et al., 1975; Lin et al., 1988; Lin et al., 1991 and Uekama et al., 1987). These reports prepared IDM inclusion complex by using different method and CDS. The

nature and the physicochemical properties of IDM products depended on the method of preparation used. Furthermore, the more amount of CDS, the more dissolution rate obtained. There are several methods to prepare the inclusion complex such as coprecipitation(Kurozumi et al., 1975, Kedzierewicz, 1990), lyophilization or spray-drying (Gandhi and Karara, 1988; Helm et al., 1991; Kurozumi, Numbu and Nagai, 1975; Lin and Kao, 1989; Lin et al., 1991; Oguchi et al., 1990), cogrinding (Çelebi and Erden, 1992; El-Gendy et al., 1986; Lin, Kao and Yang, 1988; Nakai, 1986), kneading(Nozawa and Yamamoto, 1989; Uekama et al., 1987) and the latest, sealed heating (Nakai et al., 1987, 1989; Yamamoto et al., 1991).

In this present study, the simple dispersed technique was employed to prepare IDM dispersed systems such as grinding, kneading and solvent method. The natural CDS (α-,β- and δ-CD) and β-CD derivatives,heptakis-2,6-di-O-methyl-β-CD(DIMEB) were used as carriers in these studies. As the dissolution rate was considered to be of primary interest, the objective was to increase the dissolution of IDM when incorporated with these CDS. From the preliminary study, the data showed that the dissolution of the IDM dispersed systems were improved, especially in the initial stage and also observed the change in the physicochemical properties of IDM molecules. Thus, the effect of the physicochemical properties of IDM in dispersed systems on the dissolution was also investigated. Moreover, the type of preparation, type and quantity of CDS that influenced on the dissolution and the physicochemical properties of IDM were studied, too.

For the β-CD derivative, DIMEB, claimed to has superior properties than the parent CD, β-CD such as high solubility in water

(57 g/100 ml at 25 °C) and many organic solvent such as alcohol (0.74 g/100 ml) (Duchêne, 1990b), higher solubilizing capacity, less hygroscopic that is of advantage in the case of moisture-sensitive drugs, so that DIMEB can preferably be applied for solid dosage forms (Szemán et al., 1988). There are several reports showed the increasing dissolution of drug molecule via inclusion complex or incorporated with DIMEB (Green, Miller and Guillory, 1991; Uekama et al., 1985; etc.) For IDM, DIMEB was found to be the most efficient solubilizing agent compared with hydroxypropyl-β-CD(HP-β-CD), 2,3,6-tri-O-methyl-β-CD (TRIMEB), randomly methylated β-CD (RAMEB), 3-monosuccinyl-heptakis-2,6-dimethyl derivative of β-CD (SUMEB) and β-CD when studied phase solubility diagram (Szemán et al.,1988). Among the four CDS used in this study, the DIMEB has the highest solubility in water followed by δ-CD > β-CD > α-CD, respectively. Then DIMEB was chosen to study and compare with these three natural CDS.

Purposes of investigation

1. Study and compare physicochemical properties of IDM powders that obtained by the simple dispersed techniques such as grinding,kneading and solvent method using different CDS as carriers.

2. Study the influence of physicochemical properties of IDM dispersed systems, type (α-, β-, δ-CD and DIMEB) and quantity of CDS used on the dissolution profiles.

3. Select appropriate IDM-CD systems to prepare capsule and compare dissolution profile with the corresponding powder formula.

Literature Reviews

The dissolution-absorption process of an orally administered drug can be expressed to the first approximation by the following kinetic model:

$$F \xrightarrow{k_d} GI \xrightarrow{k_a} B \qquad (1)$$

solid drug — dissolved drug — absorbed drug

where : F is drug in the orally administered dosage

k_d is dissolution rate constant

GI is concentration of dissolved drug in the gastrointestinal tract

k_a is absorption rate constant

B is concentration of drug in blood

In many cases, $k_d < k_a$ (i.e., the drug is poorly soluble in water), and therefore the dissolution rate is the limiting factor. In such cases, k_d must be increased in order to improve drug delivery. This can be done by the following methods.

1. Reduction of particle size

A decrease in the size of the particles increases the effective surface area of material in contact with the stationary layer of solvent and therefore the rate of solution increased. The enhancement of drug dissolution and absorption could further be increased several fold if a micronized product was employed.

Decrease in size or increase in effective surface area does not always result in faster dissolution rate. When the drug is hydrophobic, aggregation and agglomeration, or air adsorption may result in poor wettability and reducing the effective surface area and dissolution.

2. Application of polymorphism properties of the drugs

The phenomenon of a compound in more than one crystalline state is known as polymorphism. Different polymorphs are known to possess different physicochemical properties such as solubility, dissolution rate, density, crystal habit, melting point and stability. A more soluble polymorphic form was used to improve the bioavailability of relatively insoluble drugs. A 50-100% increase in dissolution rate can be realistically achieved through polymorphic modifications(Shefter, 1981).

3. Use of water soluble salt forms

The solubility of organic drug compounds, which are containing weak acid or base moieties, can be enhanced by the use of the salt form of the drug; the smaller the counterion, the more soluble the compound is likely to be.

4. Solubilization by surfactants

Surfactant or surface-active agents have variable effects on the dissolution and bioavailability of a drug. Drug particles

usually are hydrophobic and poorly wet by dissolution medium. In the presence of surfactant, the fine particles are wetted and the dissolution rate is enhanced. Furthermore, the solubility of a drug is improved when the surfactant is present in excess of its critical micelle concentration(CMC) but not when it is below its CMC. Singla and Mediratta developed zinc-indomethacin complex and studied the effect of sodium lauryl sulphate on the drug release kinetics from the capsule formulations. They found that sodium lauryl sulphate enhanced the percentage of the drug dissolved (Singla and Mediratta, 1988).

5. Use of highly effective disintegrants

Disintegrants produce rapid disintegration time and this can lead to increase in dissolution rate and therapeutic effectiveness of the drug. Tarimci and Çelebi studied the effect of CD polymer which is a crosslinked derivatives of β-CD, on IDM tablet formulation prepared by direct compression method. They found that CD polymer was the good disintegrating agents. The dissolution increased significantly as well (Tarimci and Çelebi, 1988).

6. Solubilization by polymerphic carriers

The incorporation of drugs into solid water soluble carriers has frequently been reported to result in an increase in the drug dissolution rate, often leading to an improvement in bioavailability. Such dosage forms are terms "solid dispersions", these being defined as "the dispersion of one or more active

ingredients in an inert carrier or matrix at solid-state prepared by the melting(fusion), solvent or melting-solvent method".

The dissolution of IDM-PVP coprecipitates have been investigated by Corrigan et al. They reported that co-spray drying IDM with up to 20% PVP gave a fused amorphous solid. Apparent solubility and dissolution studies illustrated the higher energy of IDM in these systems. The presence of PVP in the solid retarded conversion of indomethacin to a crystalline phase, the effect increasing with increasing PVP content (Corrigan, Holohan and Reilly, 1985). Somlak elucidate the ability and mechanism of enhancing dissolution of IDM systems prepared by solvent solid dispersion with mannitol,PEG 4000,PVP K 30 and SLS. All four systems, solid dispersion systems showed the greatest dissolution, followed by physical mixtures, treated IDM and pure drug. He found that IDM polymorph Form I to Form II in all solid dispersion as in treated drug, except for IDM-PVP K 30 solid dispersion which showed an amorphous form. Complex formation was also appeared in IDM-PVP K 30 and higher ratio of IDM-SLS solid dispersion. Furthermore, liquid penetration studied demonstrated that wettability increased in both solid dispersion and physical mixture (Somlak,1991).

Ford and Elliott reported the effect of particle size on the dissolution rate of IDM-PEG 6000 dispersions and the gastrotoxicity in the 10% IDM dispersions. It was apparent that an optimum size range of particles existed for maximum dissolution rates but the larger sized particles produced more gastric damage than the smaller particles (Ford and Elliott, 1985).

7. Solubilization by solid surface dispersion method

The dissolution characteristics of drug can be altered by dispersing it on the surface of an inert carrier (Shefter, 1981). The solid surface dispersion systems are achieved by two methods, deposition of the drug by solvents on solid supports (solvent deposition) and by grinding it with certain materials such as cyclodextrins or microcrystalline cellulose (frictional deposition or mechanical deposition). Monkhouse and lach used "miniscular form" to describe the state of the resulting material contained the drug in a "molecularly micronized" state on the surface of the carrier. The fast dissolution rates of these systems was found to be dependent on the weight ratio of drug to excipient. When low fractions of drug are used, the rate is maximized.

8. Solubilization by cyclodextrins and their derivatives

Cyclodextrins are well known for ability to encapsulate various molecules in the molecular state, conferring on them new physicochemical and pharmacotechnical properties, including increase solubility, bioavailability, greater stability and reduce incidence of side effects. The physicochemical characteristics and pharmaceutical uses (especially increasing dissolution) of CD and CD derivatives are present in detail.

CYCLODEXTRINS (CDS)

The α,β and δ-CDS are cyclic oligosaccharides consisting of six, seven and eight glucose units which can be obtained on a large scale from starch. The ring formed is externally very hydrophilic and relatively apolar internally. CDS were interested in the pharmaceutical sciences due to their abilities to interact with drug molecules to form complexes which can confer disirable properties on the drugs. The pharmaceutical applications of CD complexes and complexation phenomena may include (Bekers et al., 1991; Jones et al., 1984; Szejtli, 1988):

a) conversion of a liquid material into a solid product

b) masking an unpleasant taste or odour of a compound

c) avoidance of an incompatibility of uncomplexed compound with other drugs or excipients in a formulation

d) stabilisation of a compound which could otherwise be sensitive to temperature, hydrolysis, autooxidation, photodegradation etc.

e) increasing aqueous solubility and ease of emulsification of a compound of low aqueous solubility

f) enhancing the in vivo absorption and hence bioavailability of a drug with low aqueous solubility (because of an increase in dissolution rate) finally, drug-induced local irritation is reduced

g) specific reactions (such as catalysis) can be made more selective by the inclusion of specific functional groups

h) the color of a compound may be changed as inclusion usually involves a change in spectrum of the molecule

i) altering the formation rates of isomeric products

1. Preparation, structure and properties of cyclodextrins

1.1 Preparation of cyclodextrins

CDS were isolated by Villiers from the degradation products of starch and were characterised as cyclic oligosaccharides by Schardinger. They were comprised of α-1,4-linked D-glucopyranose units.

The CDS are produced by the hydrolysis and cyclisation of starch through the action of CD transglycosylase (CTG) enzyme, most usually from certain species of Bacillus microorganism e.g. *Bacillus macerans*. The preparation of CDS can be subdivided in the following main phases:

- cultivation of a microorganism producing the CTG enzyme;
- separation of the enzyme from the medium, its concentration and purification;
- enzymatic conversion of prehydrolysed starch to a mixture of cyclic and non-cyclic dextrins;
- separation of CDS from the conversion mixture, their purification and crystallization.

The biosynthesis of CDS have been discussed in detail by Szejtli (Szejtli, 1988). Chemical systhesis of CDS have not been

reported. The yields of the various types of CD are in the rank order β>α>δ-CDS and this is reflected in their relative prices.

1.2 Structure and properties of cyclodextrins

Nuclear magnetic resonance (NMR) and X-ray diffraction studies indicate the C-1 chair formation for the glucose molecules. CDS are torus shaped, as shown in Figure 1 , the secondary hydroxyl groups on the C-2 and C-3 atoms of the glucose units being located on one side of the torus, while the primary hydroxyl groups on C-6 are positioned on the opposite side of the torus.

Figure 1 The structure and numbering of the atoms of β-CD; (○— primary and —● secondary hydroxyl groups).

The hydroxyl groups on C-2, C-3 and C-6 are available as points of structural modification without danger of eliminating the "central void" which, as will be seen, is available for the accommodation of guest molecules. The interior of the torus consists of two rings of C-H groups, comprising H-3, H-5 and H-6, and a ring of glucosidic "ether" oxygen atoms. The interior of the torus of each type of CD therefore offers an environment of much lower polarity than is present in water and may be termed a "hydrophobic cavity". The cavities of CDS are slightly cone-shaped with the C-2, C-3 side more open than the C-6 side. The C-H groups comprising H-1, H-2 and H-4 are located on the exterior of the molecule. Figure 2 shows a sketch of the characteristic structural features of cyclodextrins.

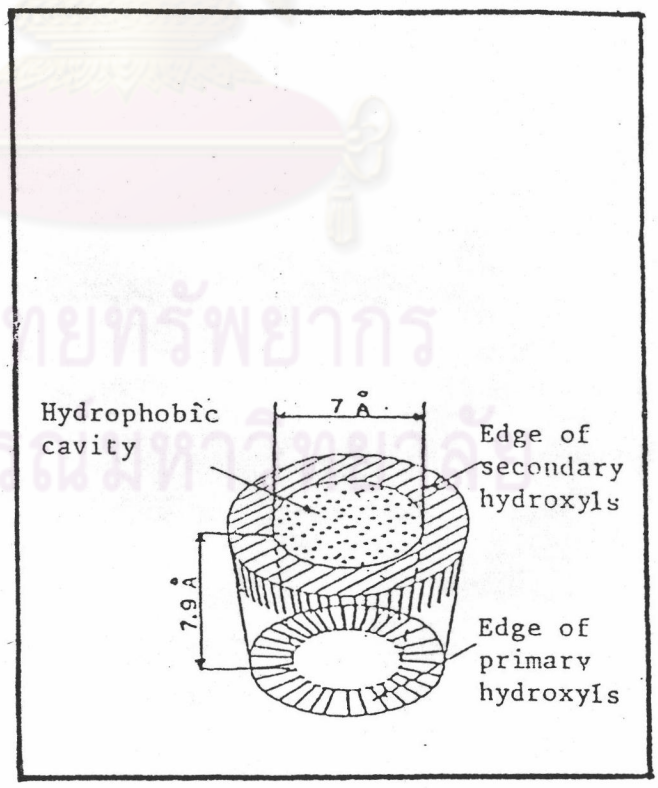


Figure 2 Functional structural scheme of cyclodextrins.

The C2-OH group of one glucopyranoside unit can form a hydrogen bond with the C3-OH group of the adjacent glucopyranose unit. These intramolecular H-bonds stabilize the CD molecule and turn the CD molecule into a rigid structure. This is the probable explanation for the observation the β-CD has the lowest solubility of all the CDS. α-,β- and ϑ-CDS are the most commonly used natural CDS. Table 1 lists some of the important physicochemical physicochemical properties of CDS.

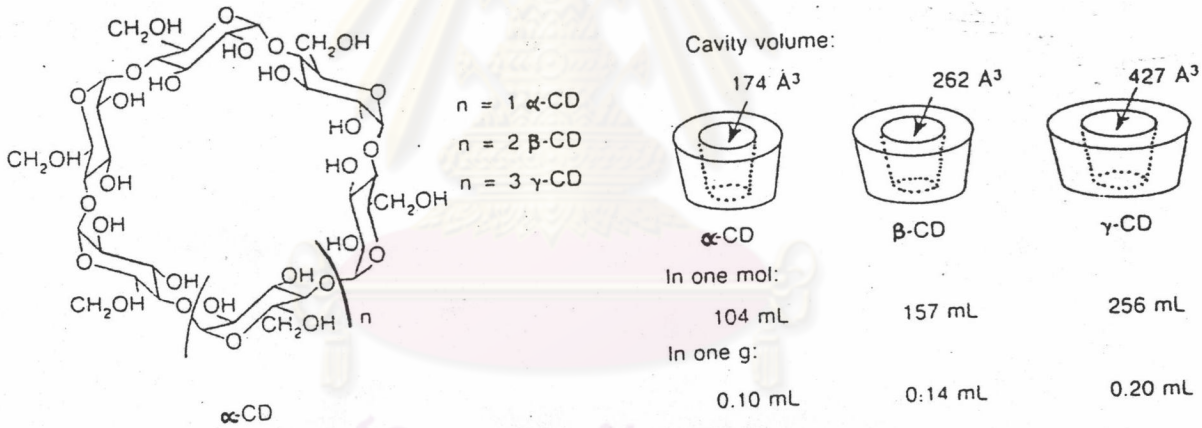


Figure 3 Structure and cavities volume of α,β and ϑ cyclodextrins.

Table 1 Physical properties of the natural CDS.

Physical properties	α	β	δ
Number of glucose residues:	6	7	8
Cavity dimensions (Å)			
– Cavity diameter :	5	6	8
– Height of torus :	7.9	7.9	7.9
– Diameter of periphery :	14.6	15.4	17.5
Molecular weight :	973	1135	1297
Aqueous Solubility [1]:	14.50	1.85	23.20
Melting point (°C):	275	280	275
pKa [2]:	12.3	12.2	12.1
Half-life of ring opening [3] (hr) :	6.2	5.4	3.0
Enzymatic hydrolysis [4]:	negligible	slow	rapid
Crystal forms (from water) :	hexagonal plates	monoclinic paralellograms	quadratic prisms

1) in grams per 100 ml water at ambient temperature

2) pKa : by potentiometry at 25°C

3) Half-life of ring opening : in 1 N HCl at 60°C

4) by Aspergillus oryzae α-amylase

2. Metabolism and toxicity of cyclodextrins

After oral administration, CDS are not hydrolysed during their transit through the small intestine, hydrolysis occurring only in the colon. Their metabolisms are more or less comparable with that of starch, but which a slower initial rate, due to the fact that CDS are totally resistance to β-amylase; which degrade only free end groups, but they can be attacked by α-amylase active inside the molecules. However, their degradation rates are quite different, α-cyclodextrin having the slowest and ϑ-cyclodextrin the fastest. It seems that α-cyclodextrin is not (or is only partially) degraded in the gastrointestinal tract. Furthermore, α- and β-CDS are poorly absorbed by the small intestine (Bekers et al., 1991; Duchêne and Wouessidijewe, 1990 a; Szejtli, 1988).

The oral administration of CDS does not result in an acute toxicity. Long-term administration leads to no significant change in organs or biological values. The consequence of the parenteral administration of CDS is completely different: the intramascular administration of β-CD results in ulcerations and its intravenous administration in nephrotoxicity and haemolytic effect. Probably due to its high water solubility, ϑ-CD is not so nephrotoxic and is less haemolytic than both α- and β-CDS.

3. Cyclodextrin inclusion compounds

CDS have the ability to form complexes with a great

variety of molecules, especially for organic molecules both in the solid state and in solution, particularly aqueous. Most of these complexes are "inclusion compounds" which are so named because the CD molecules act as "hosts" into whose cavities foreign "guest" molecules may fit. The minimum requirement for this inclusion complex formation is that the guest molecule must fit, entirely, or at least partially, into the CD cavity. In general, hydrophobic molecules rather than hydrophilic ones have higher affinity to the CD cavity in the solution.

It is possible to prepare a solid complex suitable for oral drug formulations from an organic compound of an apolar character providing the following conditions hold (Szejtli, 1991):

- the skeleton of the compound consists of more than five atoms (C,P,S,N)
- the solubility of the compound in water is less than 10 mg/ml
- the melting point of the compound is below 250 °C (a higher melting point means the cohesive forces between the molecules are too strong)
- the compound consists of fewer than five condensed rings
- the molecular weight of the compound is between 100 and 400 Da (if the molecular weight is too small, the drug content of the complex is too low; if the molecular weight is too large, the molecules do not fit inside the CD cavity).

The structure of CD inclusion complexes differ significantly in the crystalline state and in solution. In solution the guest molecule occupies in the cavity, and the whole complex is surrounded by a solvate shell of water molecules. In the crystalline state, the guest molecules can be accommodated not only in the cavity of the molecule, but also in the intermolecular cavities formed by the crystal lattice, or sandwich-like between two complex molecules. Some of the CD molecules remain unoccupied or they include water. This arrangement may result in the formation of nonstoichiometric inclusion compounds.

4. Binding force of the complexes

The interaction force for inclusion complex formation cannot be a classical non-polar binding. The CD complexes formed should be stabilized by various intermolecular forces such as:

4.1 Van der Waals interactions between the guest and hosts. The van der Waals forces include both permanent induced-dipole-dipole interactions and london dispersion forces

4.2 Hydrogen bonding between the guest and host

4.3 Release of high energy water molecules in complex formation.

4.4 Release of strain energy in the macromolecular ring of the CD.

Schematic illustration of the complexation process are presented in Figure 4 .

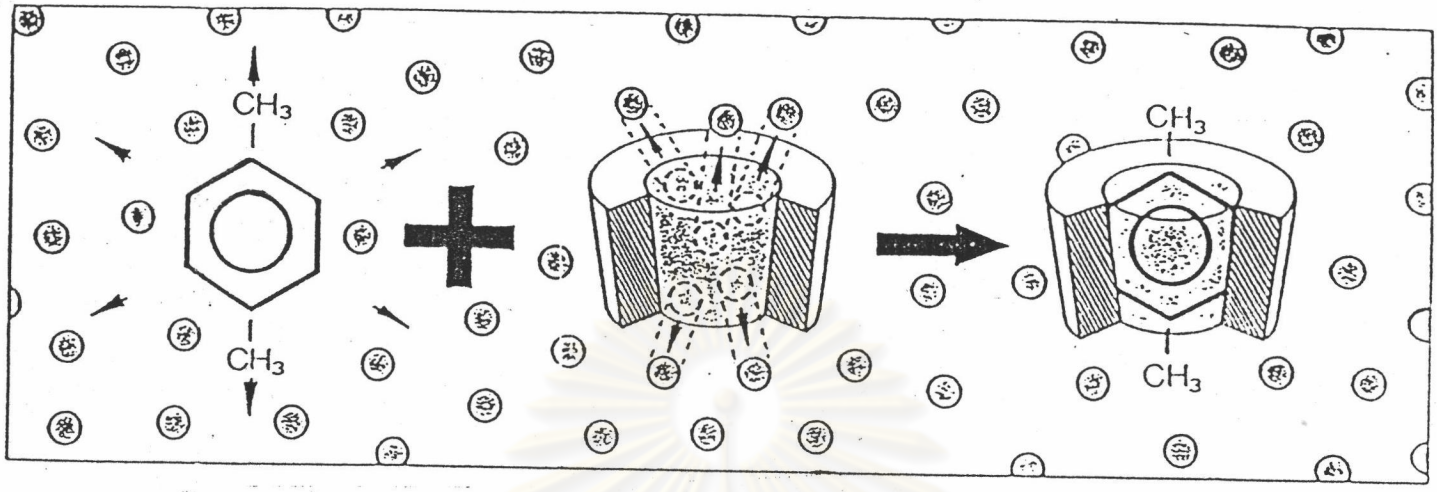


Figure 4 Schematic illustration of the complexation process. (The circles represent water molecules.)

The outer surface of the β-CD ring is hydrated, but the water molecules in the ring cavity are in an energetically unfavourable position because of the nonpolar surface of the cavity. The potential guest molecule, here P-xylene, repulses the water molecules. The result of the complex formation is that the nonpolar side of the guest molecule penetrates into the nonpolar cavity, thereby establishing an energetically favourable nonpolar-nonpolar interaction.

5. Preparation of solid cyclodextrin inclusion compounds

The method used for the preparation of CD inclusion compounds depends on the properties of the guest molecules:

5.1 In the case of a water-soluble active ingredient, the guest product is added to a saturated aqueous solution of CD, and agitation for several hours or even days, until spontaneous precipitation of the inclusion is achieved. Sometimes precipitation does not occur spontaneously, and it is necessary to cool the medium or evaporation by freeze-drying or spray-drying (Gandhi and Karara, 1988; Helm et al.,1991; Kurozomi, Nambu and Nagai, 1975; Lin and Kao, 1989; Lin et al., 1991; Oguchi et al.,1990).

5.2 When the active ingredient is highly water insoluble, it is necessary to use an organic solution of the active ingredient, which is poured, under agitation, into an aqueous solution of CD. Precipitation is obtained either spontaneously or by evaporation.

Two procedures are suitable for the laboratory preparation of cyclodextrin inclusion complexes. For the industrial scale, the grinding (Çelebi and Erden, 1992; El-Gendy et al.,1986; Lin,Kao and Yang, 1988; Nakai, 1986) and kneading process will be used.

5.3 Details of the kneading procedures are : adding the active ingredient to a slurry of CD, and kneading throughly to obtain a paste, which is then dried. The product is washed with an organic solvent to remove the free active ingredient mixed with the inclusion compound. This method, easy for industrial applications, is far from being recommended for the obtention of a pure inclusion (Nozawa and Yamamoto, 1989; Uekama et al.,1987).

5.4 Heating in a sealed container is the new method for preparing inclusion compound but it is practically restricted to sublimable guests such as menthol and benzoic acid (Nakai et.al., 1987, 1989; Yamamoto et al.,1991).

6. Phase solubility Analysis

Organic compounds, such as drugs, which are sparingly soluble in water, frequently display an increased aqueous solubility in the presence of CD. This is due to the formation of water soluble complex between the drug and the dissolved CD. The complexation equilibrium lowers the thermodynamic activity of the dissolved drug. Consequently, more drug dissolves until the activity of the free drug, which is in chemical equilibrium with the complex, becomes equal to the thermodynamic activity of the pure solid drug. Phase solubility analysis is used to determine the relationship between the total concentration of dissolved drug and the concentration of added CD. This technique reveals both the stoichiometry of complex formation and the stability constant (or formation constant, K_f) of the complex.

The solubility of drug molecules can often easily be measured, and phase solubility analysis, as applied by Higuchi and Connors (Higuchi and Connors, 1965), then enables the formation constant of the complex to be readily determined. Since CDS do not absorb in the UV the total concentration of a drug in solution is often determined by UV spectrophotometry.

Several different phase diagrams may be obtained from systems which form complexes. These are examined in some detail by Higuchi and Connors. They have divided the systems into two major classes: type A or type B diagrams.

The type A phase diagram, shown in Figure 5 , is obtained for systems in which the complex formed is soluble and does not form a precipitate regardless of the amount of ligand added. This can be subdivided according to the detailed nature of the phase diagram obtained. The A_L is obtained when exhibits a linear relationship between S_t and L_t. The A_P diagram, which shows a positive deviation from linearity, is obtained when the complexes formed contain more than one molecule of ligand. As the ligand concentration increases the contribution of the higher order complexes increases. The remaining A-type diagram, A_N, exhibits a negative deviation which represents a decreasing dependence on ligand added at higher ligand concentrations. This type is the least frequently encountered system, and its occurrence may be explained on the basis of self-association of the ligand at high concentration.

The type B diagram (see Figure 6) is obtained for those systems in which the complex actually precipitates from the solutions when the concentration of ligand exceeds some critical values. If the complex exhibits some solubility, the diagram shows an initial rise in $[S]_t$ and the diagram is said to be a B_s diagram. If the complex is insignificanty soluble relative to the inherent solubility of the substrate, then the system gives rise to the B_i diagram.

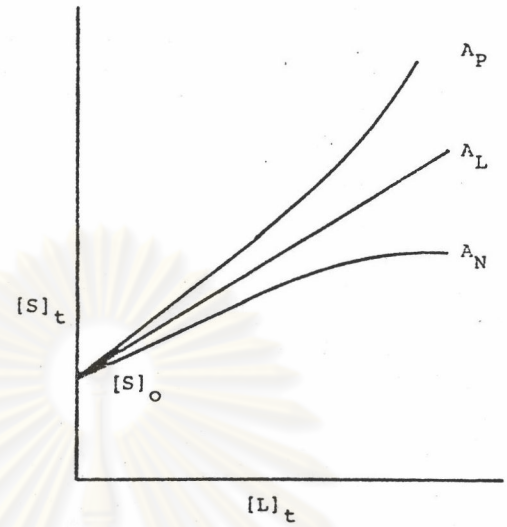


Figure 5 Schematic representation of the A-type phase diagrams ($[S]_t$ = the concentration of the total substrate in the solution and $[L_t]$ = the concentration of the total added ligand)

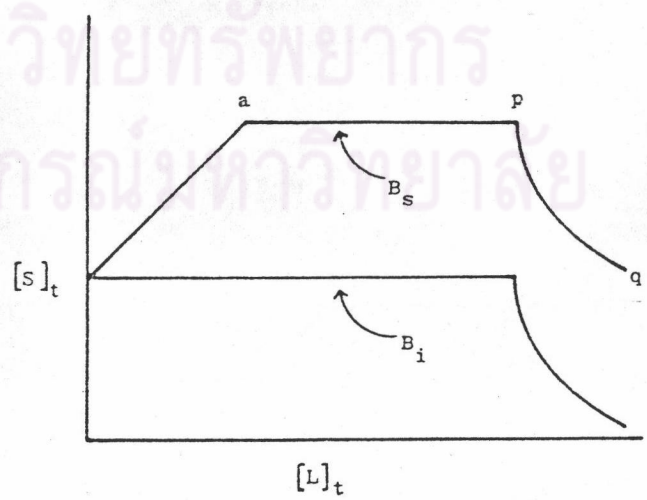


Figure 6 Schematic representation of the B-type phase diagrams.

7. Method used for investigating inclusion compounds

7.1 Detection of inclusion complexation in the solid state

7.1.1 Infrared spectra

Complex formation may be proved in some cases by IR studies, but this method is of limited use. This is due to the bands of CD representing the majority of the complex being hardly influenced by complex formation but some investigations showed the changing of the chemical shift and the broadening peak(Szejtli, 1988). In general, however, no change due to complex formation can be observed.

7.1.2 X-ray Diffraction

Liquid guest molecules do not produce diffraction patterns, if however there are differences from that of uncomplexed cyclodextrin, complex formation can be considered probable. In the case of non-volatile liquid guest molecules, X-ray powder diffraction is very useful. When the guest component is a solid substance, a comparison of the diffractograms of the assumed complex with a mechanical mixture of the guest and cyclodextrin (crystallised under identical conditions as the assumed complex) has to be made. When the diffractograms are different, i.e. the characteristic peaks of one or other of the components disappear, and new ones appear as a result of the complexation experiment, complex formation is very

probable. In the case of small guests the crystal structure may be identical with that of the cyclodextrin hydrate (Szejtli, 1988).

7.1.3 Thermo-analytical methods

Thermo-analytical methods determine whether the guest substance undergoes some change before the thermic degradation of CD. The change of the guest may be melting, evaporation, decomposition, oxidation or polymorphic transition. This method consists of many techniques, such as (a) thermal analytical system (TAS), (b) thermo evolution analyzer (TEA), (c) differential scanning calorimetry (DSC), (d) thermogravimetry (TG), and (e) differential thermal analyzer (DTA) which was used to confirm in this study.

7.1.4 Thin layer (TLC) and paper chromatography (PC)

7.1.5 Scanning electron microscopy

7.1.6 Wettability and dissolution tests

7.2 Detection of inclusion complexation in solution

7.2.1 Spectroscopic methods

This method consists of Ultraviolet/Visible(UV /VIS) spectrosccpy, Fluorescence spectroscopy, Circular dichroism (CD)

spectroscopy, Electron spin resonance (ESR) and Nuclear magnetic resonance (NMR) spectroscopy or ^{13}C-NMR may be used in confirming the inclusion complexes.

7.2.2 pH-potentiometric titration

7.2.3 Electrochemistry (Polarography and Conductivity)

7.2.4 Microcalorimetry

7.2.5 Solubility method

7.2.6 Surface tension techniques

8. Cyclodextrin derivatives

Natural CDS can be modified for many different purposes, for example to improve the low aqueous solubility of β-CD or to decrease the toxicity in parenteral applications. This characteristic has been obtained by alkylation of the hydroxyl groups (methyl-, hydroxypropyl- and also hydroxyethyl-cyclodextrins, by substitutions of primary hydroxyl groups by saccharides (glycosyl- and maltosyl-cyclodextrins), or by polymerization of cyclodextrins.

8.1 Methylated cyclodextrins

Some of the physicochemical properties of methyl CDS

are reported in Table 2 ,which shows the increased water solubility of methyl CDS. In the case of β-CD, the dimethylated derivative is more soluble than the trimethylated one. By comparison with the original CDS, the surface tension decreases for all derivatives, and their stabilities in acidic conditions (half-life of ring opening, 1.0 N HCl at 60 °C) increases for dimethyl derivatives and decreases for trimethylated ones.

Methyl CDS are not only highly soluble in water, but they are also soluble in organic solvents. For example, in ethanol, β-CD has a very poor solubility of 0.05 g/100 ml, although its dimethyl derivatives is 15 times more soluble(0.74 g/100 ml). However, their solubility decreases with an increase in temperature(Duchêne and Wouessidjewe, 1990b).

When methyl-CDS administered by the oral route, they are resistance to the bacterial hydrolyses of the gastrointestinal tract and are eliminated with faeces. They exhibit higher haemolytic effects than does β-CD itself (Jodál,Nánási and Szejtli, 1988; Szatmari and Vargay, 1988).

Table 2 Physicochemical properties of cyclodextrins and their methylated derivatives.

Molecule	Number of Glucose Units	Molecular Weight (D)	Cavity Diameter (Å)	Melting Point (°C)	Solubility in Water at 25 °C (g/100 mL)	Water Content (%)	Surface Tension (mN/m)	Half-Life 1 N HCl (h)
α-CD	6	973	5	275	15	10	71	6.2
Dimethyl α-CD	6	1141	5	260–264	—	—	65	12.6
Trimethyl α-CD	6	1225	3–6	205	—	<1	54	3.0
β-CD	7	1135	6	280	1.85	13	71	5.4
Dimethyl β-CD	7	1331	6	295–300	57	1	62	8.5
Trimethyl β-CD	7	1429	4–7	157	31	<1	56	1.7
γ-CD	8	1297	8	275	23	16	71	3.0
Dimethyl γ-CD	8	1521	8	255–260	—	—	60	4.0
Trimethyl γ-CD	8	1633	5–9	135	48	—	56	1.2

8.2 Hydroxyalkylated cyclodextrins

Example of these derivatives are 2-hydroxypropyl-β-CD and hydroxyethyl-β-CD. They have much higher aqueous solubilities than natural CDS (> 50%). As in the case of methyl-CDS, they are not hydrolysed by the gastrointestinal amylases. Also the haemolytic activities of hydroxyalkylated CDS are less than that of natural CDS. This means that they may be useful in intravenous (i.v.) and other parenteral preparations.

8.3 Branched cyclodextrins

Various branched CDS have been described, such as glucosyl, maltosyl and glucopyranosyl α- and β-CDS, and diglucosyl,

dimaltosyl and dipyranosyl-β-CDS. All are more water-soluble than β-CD. The complexation abilities of branched CDS and their parent CDS appeared to be almost the same. However, the enhancement of solubility of poorly water-soluble drugs by branched CDS is much more marked than by their parent CDS.

8.4 Other cyclodextrin derivatives

- Ethyl CDS : ethylation of CDS reduces their water solubility in proportion to the degree of substitution. They are used in sustain release preparations (Hirayama et al.,1988).

- Carboxymethyl ethyl CDS: they are characterized by a pH-dependent solubility : below pH 2.5, solubility is almost constant (1 to 1.5 g/100 ml), it then increased sharply above pH 4 (10 g/100 ml) and, at pH > 6, the product is freely soluble. The product is proposed for preferential drug release in the intestinal fluid with a very slight release in the gastric fluid.

- Cyclodextrin polymers : they are substances containing at least two CD units. These high molecular weight derivatived of CDS may be water-soluble and moderately swelling or insoluble and strongly swelling. Polymer with high molecular weights can be used as tablet excipients because they have good disintegrating properties (Fenyvesi, Shirakura et al.,1984; Fenyvesi, Takayama et al., 1984).

Table 3 and 4 illustrate which CDS are recommended for certain drug formulation purposes and some CD-containing drugs that are marketed already, respectively (Szejtli, 1991).

Table 3 Choice of cyclodextrins for drug formulation.

Cyclodextrin	Most Recommended Formulation	Remarks
α	Parenteral (limited to intraarterial infusion)	E.g., in intraarterial infusion of PGE_1–α-cyclodextrin. Use is restricted to small molecules or molecules containing slim side chains.
β	Oral (tablets)	E.g., in piroxicam–β-cyclodextrin to improve bioavailability and in garlic oil–β-cyclodextrin to improve stability. The useful properties and low cost of β-cyclodextrin mean that it will be used extensively in solid dosage oral formulations.
γ	Parenteral	In most cases, parenteral formulations can be made using modified cyclodextrin. However, for some extremely large drug molecules (e.g., macrolide antibiotics or large, substituent-bearing steroids), it will be the most promising choice.
Methylated cyclodextrins (e.g., DIMEB)	Parenteral (limited to small doses or highly diluted infusions); oral (in controlled release); topical	These are the most hydrophobic cyclodextrins and are the best solubilizers, but they also have the strongest haemolytic activity. Recommended for drugs that are highly hydrophobic or extremely sensitive to humidity.
Hydroxypropyl β-cyclodextrin (HPBCD)	Parenteral	The first choice for parenteral formulations. Approval and marketing is expected within two years.
Hydroxyethylated β-cyclodextrin (HEBCD)	Parenteral	Could be used instead of HPBCD but displays no specific advantage over the latter.
Branched cyclodextrins	Oral	One or two glucoses or malloses are enzymatically attached to the cyclodextrin ring to form these cyclodextrins. Very heterogeneous, amorphous, and nonhaemolytic. Eventually these cyclodextrins will be used in oral and topical formulations.
Ethyl- and ethyl-carboxy-methyl-cyclodextrins	Oral	Complexes made with these cyclodextrins are poorly soluble, and so sustained release can be achieved. Nothing is yet known about their toxicology.
Dihydroxypropyl cyclodextrins and soluble neutral and ionic cyclodextrin polymers	Oral; topical	Less haemolytic than HPBCD, but nothing is known about their toxicity. The soluble polymer cyclodextrins slow down diffusion of the drug. In many cases, these cyclodextrins are better solubilizers than HPBCD.

Table 4 Approved and marketed cyclodextrin drug complexes.

Complex	Trade Name	Formulation	Indication	Company/ Country
PGE_1–α-CD	Prostavasin	Intraarterial infusion	Vasodilator	Ono, Japan; Schwartz, FRG
PGE_2–β-CD	Prostarmon-E	Sublingual tablet	Initiation of labour	Ono, Japan
Piroxicam–β-CD	Brexin Cicladol	Tablet and suppository	Analgesic, anti-inflam-matory	Chiesi, Italy; Master-pharma, Italy
Garlic oil–β-CD	Xund Allidex	Dragees	Antiathero-sclerotic	Bipharm, FRG; Fov.Gyógy-szert. K., Hungary
Benexate–β-CD	Ulgut Lonmiel	Capsule	Antiulcerant	Teikoku, Japan; Shionogi, Japan
Iodine–β-CD	Mena-Gargle	Gargling	Throat disinfectant	Kyushin, Japan

9. The application of CDS and their derivatives inclusion complexes concerning the enhancement of drug dissolution and bioavailability

Physicochemical properties of the guest molecule may be changed by CD inclusion complexation. However, the greatest interest in CDS arises from their capacity to modify the solubility of the drug. Substances which are sparingly soluble in water can be made more soluble and the rate of dissolution may be enhanced by the addition of CDS. This can lead to a higher pharmaceutical availability and enhanced bioavailability after oral administration. Several papers have been published that describe the extensive application of CD complexation to enhance to solubility, dissolution rate and bioavailability of slightly soluble drugs (Çelebi and Erden, 1992; Gandhi and Karara, 1988; Kedzierewicz, Hoffman and Maincent, 1990; Marques, Hadgraft and Kellaway, 1990; Mura et al.,1988; Nakai, 1986 Otero-Espinar et al.,1992; Seo et al.,1983; Tasić, Jovanović and Djuvic,1991; Weiszfeiler, Stadler-Szóke and Szejtli, 1988; etc.). Since many derivatives of CDS were synthesized to improve the solubility of β-CD: eg. methylated CDS, hydroxyalkylated CD etc. Many researchers found that with increased water solubility of the derivatives the complex-forming capacity could be increased in comparison with the starting compound, too (El-Gendy et al., 1986; Green, Müller and Guillory, 1991; Helm et al., 1991; Loftsson et al; 1991; Uekama et al., 1985 etc.).

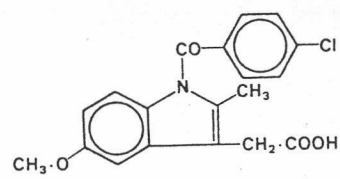
10. The application of CDS as filler-binder in dosage formulation

Shangraw et al. characterized the tableting properties of a number of commercially available β-CD and found that compactibility was excellent but varied by source. Inclusion complexes spontaneously formed during wet granulation processing. Furthermore, the presence of β-CD in a formulation will enhance dissolution of drugs of low water solubility even if the drug is not in the form of an inclusion compound. Thus, β-CD could be useful as a tablet and capsule filler (Manna,Giordano and Gazzaniga,1990; Shangraw,Pande and Gala,1990).

β-CD was evaluated as a direct compression vehicle either singly or in blends with spray-dried lactose for preparing tablets containing either phenobarbitone, diazepam, prednisolone or spironolactone. It was found that β-CD and its combinations produced tablets having very good mechanical properties and higher dissolution rates. Moreover, the optimum formulation was found to vary from one drug to another depending upon it nature, dose and molar ratio of inclusion complex with β-CD (Elshaboury,1990).

Indomethacin (Borka, 1974; Brien, McCauley, Cohen, 1984)

The molecular structure of Indomethacin is shown below.



The empirical structure is $C_{19}H_{16}ClNO_4$ with molecular weight 357.81. IDM is pale yellow to yellow-tan, crystalline powder that is odorless or almost odorless with a faintly astringent taste. It can exist as several crystalline forms. The various polymorphs and the corresponding melting point are tabulated in Table 5 .

Table 5 Melting point of Indomethacin polymorphs.

Form	Melting point(°C)
Form I (type δ)	160-161.5 160 158
Form II (type α)	154.5-155.5 154 152
Form III	148
Form IV	134
Type β	158-160.5
Amorphous	67

Form I is the highest melting and lowest solubility polymorph and is, therefore, the thermodynamically stable crystalline modification of indomethacin. However, from a practical view, both Form I and II are equally biologically available and active.

The following solubility data have been reported in Table 6 .

Indomethacin is a non-steroidal, anti-inflammatory agent(NSAID) with antipyretic and analgesic properties. It has been used effectively in the management of patients with moderate to severe rheumatoid arthritis, ankylosing spondylitis, osteoarthritis, acute painful shoulder (bursitis and/or tendinitis) and acute gouty arthritis. The recommended daily dose was 50 to 200 mg.

Stability : Indomethacin powder and formulated products exists for at least five years at room temperature. Exposure to strong direct sunlight induced an increase in the color of indomethacin; however, degradation is slight. The degradation of indomethacin occurs primary via hydrolysis of the amide moiety. In both acid and basic solutions, indomethacin undergoes acidic and alkaline hydrolysis to p-chlorobenzoic acid and 2-methyl-5-methoxy-indole-3-acetic acid.

Stabilization method : Besides using for solubilization, nonaqueous solvents were used to stabilize IDM solutions. Amber-colored containers and antioxidants have been used to decrease the photolytic degradation of IDM. Furthermore, use of TiO_2 in the gelatin films of IDM capsules reduces photodegradation of solid indomethacin (Connors, Amidon and Stella, 1986).

Table 6 Solubility data of IDM from various solvents.

Solvent	Temperature (°C)	Solubility
Water	25	0.40 mg/100 ml[a]
	25	0.52 mg/100 ml[b]
	25	0.88 mg/100 ml[c]
	RT	Practically Insoluble
Phosphate Buffer pH 5.6	25	3 mg/100 ml[a]
	25	5 mg/100 ml[b]
Phosphate Buffer pH 6.2	25	11 mg/100 ml[a]
	25	16 mg/100 ml[b]
Phosphate Buffer pH 7.0	25	54 mg/100 ml[a]
	25	80 mg/100 ml[b]
Ethyl alcohol (95%)	RT	1 in 50
Chloroform	RT	1 in 30
Ether	RT	1 in 45
Methanol	25	32 mg/gm
Benzene	25	4 mg/gm
n-butanol	25	19 mg/gm
sec-butanol	25	27 mg/gm

Note : a = Form I

b = Form II

C = Form III

Alkaline hydrohysis of indomethacin was stabilized by using cosolvent, especially PEG 400, that could reduce the dielectric constant of the mixture or by adding nonionic or ionic surfactants which incorporate the indomethacin into micelles. The order of surfactants in increase in the stability of indomethacin was : polysorbate 80 > sodium lauryl sulfate > benzalkonium chloride > cetrimonium bromide (Suleimann and Najib, 1990).

Hamada et al. investigated the interaction of indomethacin with α- and β-CD and found that β-CD stabilized aqueous solutions, whereas α-CD tended to produce the opposite effect (Hamada, Nambu and Nagai, 1975). Müller and Brauns found that IDM can be stabilized with hydroxyethyl-β-CD with solution investigated were not physically stable during the investigated period of 6 months, due to microbial contamination (Müller and Brauns,1985). Moreover, the β-CD derivatives inhibited the hydrolysis of IDM in phosphate buffer pH 7.4 more effectively than the parent did (Backensfeld et al.,1990).

Solubilization method : Hamada et al. reported the interaction of α- and β-CD with several non-steroidal antiinflammatory drugs, including IDM, and found that the dissolution rate of drug increased with β-CD, while not with glucose (Hamada, Nambu and Nagai,1975). Kurozumi et al. reported that the freeze-drying method was successful in obtaining the inclusion compound of all the tested, NSAIDS including IDM with β-CD. Some of the drugs also formed inclusion complex with α-CD. The freeze-drying method gave a good yield of inclusion compounds comparable to the usual coprecipitation method

(Kurozumi et al.,1975). Myles et al. found that IDM forms inclusion complexes with α-, β- and δ-CD by using phase solubility and NMR. The stability constants for the complexes are found to vary in the rank order β≃ > α- >> δ-CD. An inspection of the computer graphics space filling models for the CD complexes shows that the CD cavities are almost completely filled by the guest in the α- and β- complexes, but only partially occupied in the δ- complex (Myles et al.,1990).

The complexes of indomethacin/β-CD or its derivatives,hydroxypropyl-β-CD can be obtained by using different methods : kneading, spray-drying, neutralization followed by freeze-drying method. Lin et al. reported that the nature of the end products depend on the method of preparation. The kneading method did not lead to a real inclusion while the neutralization method did. In any case, the complexes obtain may be of great value as rapid dissolving forms of IDM in water. The author found that at 3 mins, nearly 95% of IDM is dissolved from the kneaded complex, compared with only 39% from the physical mixture (Lin et al.,1991).

Inclusion complexes of drug (acetaminophen, indomethacin, piroxicam, warfarin) with β-CD prepared by using a spray-drying method. It was found that the spray-drying technique could be used to prepare the amorphous state of drug inclusion complexes. The enhanced dissolution rate of spray-dried products might be attributed to the decreased particle size, the high-energetic amorphous state and inclusion complex formation (Lin and Kao, 1989).

Uekama et.al.studied on complex formations of IDM with three water-soluble CD-EP polymers (α-CD.EP,β-CD.EP and δ-CD.EP) in aqueous solution and in solid phase. Through the binding to CD.EP polymer, the solubility and dissolution rate dissolution rate of IDM increased significantly in the order of α-CD.EP > β-CD.EP > δ-CD.EP, compared to those of parent CDS. The rapidly dissolving form of IDM-α-CD.EP complex was found to increase the serum levels of drug after oral administration to healty men. Among the three CD.EP polymers, α-CD.EP seemed to be particularly useful for improving the oral bioavailability of IDM (Uekama et al.,1987).

Szemán et al. compared the solubilizing capacity of some highly water soluble β-CD derivatives on several drugs and IDM. They found that the methylated β-CD, especially DIMEB was the potent solubilizers for all studied guest molecules. Furthormore, the crystalline methylated CDS are non hygroscopic, therefore these derivatives are of advantage in case of moisture sensitive drugs and can preferably be applied for solid oral dosage forms (Szemán et al., 1988).

Grinding was widely performed as a means to reduce the particle size of powders. It has been found that grinding not only causes changes in the molecular behavior (such as phase transition of polymorphs, crystallinity and chemical reaction rate in solid state) of the ground drugs by the addition of additives but also improves the molecular interaction between drugs and additives in the ground mixtures. Lin et al. studies grinding effect on some physicochemical

properties of drugs (acetaminophen, warfarin, indomethacin, diazepam and hydrocortisone acetate) by adding β-CD. It was found the crystallinities of drugs decreased with increasing grinding time and became amorphous or nearly amorphous. The result indicates that only acetaminophen formed an inclusion complex, although all five drugs interacted with β-CD in water. This dissolution rate of drugs form the ground mixtures was shown to be higher than that of ground drug, crystalline drug or physical mixture due to the decrease in crystallinity. Furthermore, β-CD has a surfactant-like property which can reduce the interfacial tension between a water-insoluble drug and a dissolution medium, leading to a higher dissolution rate (Lin and Kao, 1988; Nozawa and Yamamoto, 1989).