CHAPTER IV

DISCUSSION AND CONCLUSION

DISCUSSION

IDM prepared by various dispersion methods with cyclodextrins as carrier illustrated faster dissolution than its corresponding drug such as grounded drug, kneaded drug, treated drug and pure drug. The method of preparation, quantity and type of cyclodextrin influenced the differences in drug dissolution profiles. Furthermore, changing in physicochemical property of IDM in some dispersed systems also gave the markedly increasing in dissolution.

It is important to identify the physicochemical properties of the drug, when it incorporated with cyclodextrin, as they may have different pharmaceutical properties, especially in solubility and dissolution. SEM, IR, DTA and X-ray diffraction were used for evaluating of such properties and explaining the differences dissolution profiles obtained. The SEM would differentiate crystal shape, habit and appearance. X-ray method would definitely confirm the different molecular configuration within the solid while IR and DTA would provide useful additional supported information.

The method of preparations of this present study were grinding, kneading and solvent method. The dispersion methods were

simple and ease to prepared; moreover this may be used in the manufactures. The objective of this study was to obtain the formulation that gave the increasing in dissolution for developing IDM powder. Furthermore, to study the influence of dispersion methods, type and quantity of cyclodextrins and physicochemical properties of dispersion systems on dissolution rate of IDM powders and capsules.

Cyclodextrins are valuable material that could formed inclusion complex with many organic compounds and offer desirable properties utilized in pharmaceutical fields. Natural CDS are α-, β- and δ-CD. The solubility in water are 14.5, 1.85 and 23.2 g/100 ml, respectively. The most useful natural CDS is β-CD because it has a suitable cavity size to include the majority of drug molecules; moreover it can produced in the large scale with a reasonable price. However, β-CD is the least soluble, many derivatives of β-CD are more interesting. In this present study,the DIMEB is used to compare with the natural CDS and its solubility in water is 57 g/100 ml.

IDM is practically insoluble drug and known to exist in a number of polymorphic states. Borka revealed that IDM have four polymorphs, Form I-IV (Borka, 1974). They were prepared by various organic solvents. The IR spectra of IDM polymorphs are presented in Figure 74 . The IR spectra are different in each polymorphs at carbonyl stretching region. This suggests the involvement of the C-O groups in different types of hydrogen bonds in building up the crystal lattice of the four modifications. In this Figure, the IR spectra of Form III is not presented because none of the

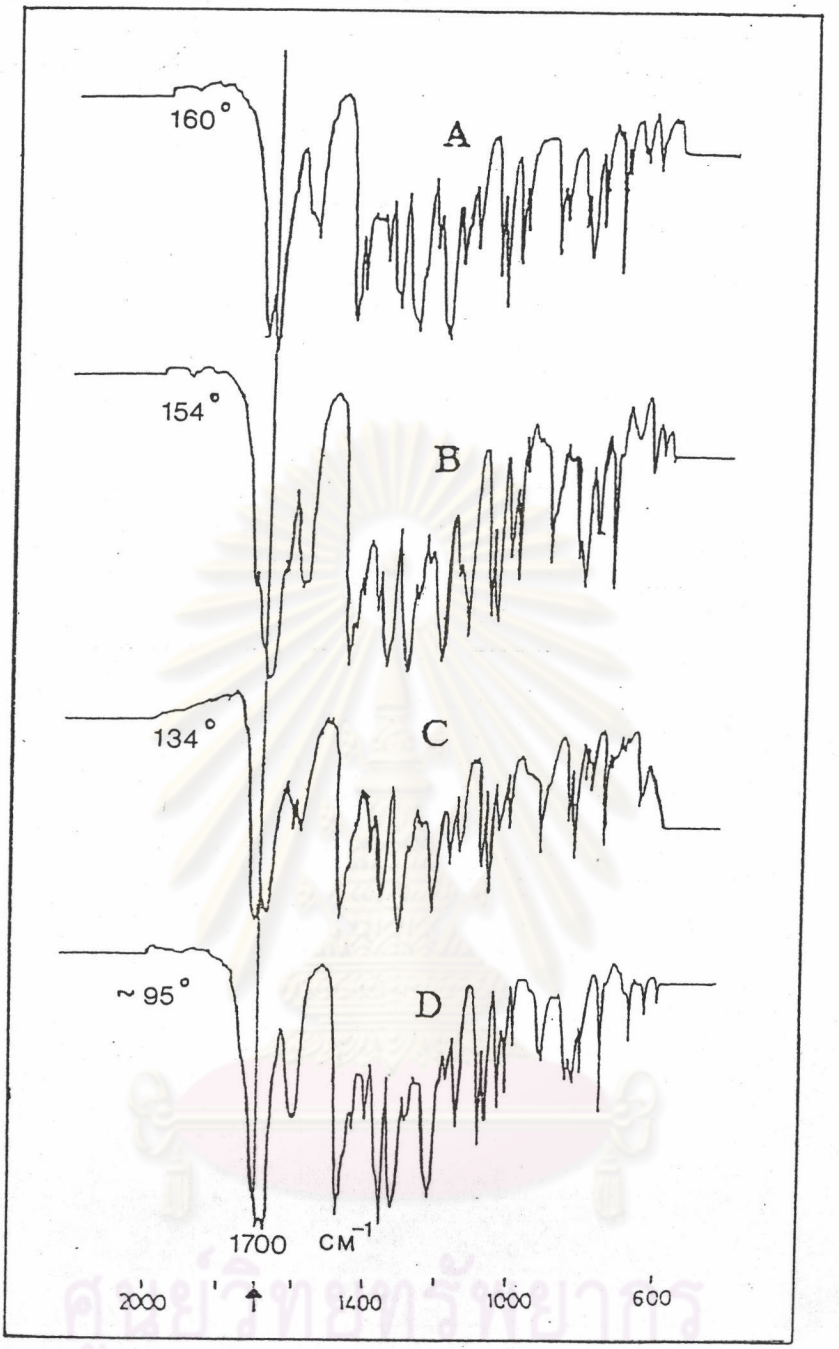

Figure 74 The IR spectra of four polymorphic modifications of indomethacin (KBr disc). From the top : A: Form I; B: Form II; C: Form IV and D: the solvent-containing form.

solvents studied in his work yielded this Form and no preparative method was found to collect in pure Form, too.

Dissolution studies

The dissolution profiles of IDM were ranked as followed : grounded drug > pure drug > treated drug > kneaded drug. IDM powder exhibited poor dissolution owing to its hydrophobicity and occurrence of clumping together of the drug particles in the dissolution medium. The grounded drug gave the highest dissolution because the particle size was reduced hence increased in surface area and dissolution according to Noyes Whitney equation. Kneading drug with water made the particles to agglomerate and reduced in surface area to contact with dissolution medium. Dissolution of treated drug was more soluble than kneaded drug at the same size range of study. This was explained by the altering in polymorph of IDM from Form I to Form II. This changing was evidenced by the particle appearances, the DTA thermograms, IR spectra and X-ray diffractograms. When comparing between pure drug and treated drug, it was observed that dissolution profile of pure drug is better. This may be explained that pure drug had various particle size ranges especially small particles (from SEM) while treated drug exhibited the agglomeration of fine needle crystalline powder. This would reduced the surface area and caused the dissolution of treated drug lower than that of pure drug.

1. Grounded mixtures

Grinding is used in order to produce the drug powder exhibiting a high surface area available for dissolution and also to ensure homogeneity of drug to provide dosage uniformity. Furthermore, it had been reported that grinding not only reduces the particle size but also caused the changes in the molecular behavior such as phase transition of polymorphs, crystallinity, and chemical reaction rate in solid phase.

The dissolution profile and time for 80% released of IDM showed that when increase the quantity of CDS in grounded mixture, high dissolution profile and less time for T80% was obtained. It was observed that DIMEB grounded mixtures gave the highest dissolution profile followed by $\delta \simeq \alpha > \beta$. These may due to the solubilizing effect of DIMEB owing to its solubility in water is very high, in addition the phase changed of IDM from polymorph Form I (0.4 mg/100 ml) to Form III (0.88 mg/100 ml) (Borka, 1974) even at the small ratio of IDM : DIMEB and the particle size reduction caused by grinding. Moreover, the inclusion complex between the IDM and DIMEB possibly occur in the dissolution medium thus causing the improve in dissolution (Szemán et al., 1988). Grounded mixtures of α- and δ-CD gave the higher dissolution profile than that of grounded drug and pure drug because the solubilizing effect of CDS due to CDS molecules were more soluble in dissolution medium than drug molecules. Furthermore, size reduction caused by grinding also caused the high surface area

hence increased in the dissolution. In the case of β-CD, at the ratios of 1:0.5 - 1:3, they exhibited lower dissolution than other CDS, afterthat when the ratios increased more than 1:3, the higher dissolution was obtained. This may be the effect of the lowest solubility of β-CD and no interaction between β-CD and IDM at the ratio lower than 1:3. The increasing in dissolution at the higher ratios (> 1:3) was supported by DTA thermograms and solubility test which indicated that the partial inclusion complex of β-CD and IDM was presented in the grounded mixtures and in the dissolution medium.

2. Kneaded mixtures

The dissolution profiles of kneaded mixtures were faster than that of the control; pure drug and kneaded drug. It could be stated that kneading method help improvement of dissolution characteristics. Similar results were previously reported by many researchers (Lin et al., 1991; Nozawa and Yamamoto, 1989; Uekama et al., 1987). This was mainly due to improvement of wettability of the drug powder by changed the surface characteristic. This effect was enhancing in the hydrophilicity and solubility of the drug.

The dissolution of kneaded mixtures were ranked as follow : DIMEB >δ-CD > β-CD > α-CD. The highest dissolution of DIMEB kneaded mixtures might be due to the better solubilizing effect, the transformation of polymorph I to polymorph III (DTA thermograms) and size reduction (SEM). Furthermore, during the

preparation of the DIMEB kneaded mixtures, the products obtained were yellow distinct from other kneaded products. Therefore, the changing in color may be owing to some interaction between DIMEB and IDM which could be inclusion complex during the preparations. And the increasing of IDM dissolved in DIMEB solution from phase solubility diagram was observed which indicated the occurrence of the inclusion complex in dissolution medium. For δ-CD kneaded mixtures, they exhibited similar dissolution as compare to DIMEB, especially at the ratio above 1:2, this result may be due to the reason as previously mentioned for δ-CD grounded mixture. However, δ-CD kneaded mixture gave higher dissolution than that of grounded mixture because of some interaction of δ-CD and IDM may be occur due to the change of color of the product (pale yellow) during the preparation. This may be due to partially inclusion complex of the mixture. However, the IR spectra, DTA, thermograms did not change except for SEM showed very small particle size of IDM particles and some change in environmental of drug molecules, solubility test gave a little improvement of solubility of IDM in the presence of δ-CD. Further investigation must be needed to obtain the explanation of this phenomena.

The enhanced in dissolution rate of β-CD is probably due to either the improved wettability and increased solubility which arose from interaction between the drug and β-CD molecules during preparation and in the test solution. DTA thermograms showed the small endothermic peak at 217-218 °C, this indicated the partially inclusion complex in the systems. The result was in accordance with

Lin et al., they reported that kneaded method could not obtained the true inclusion compound; however, it gave the high dissolution compared with the pure drug and would be useful in industrial manufacture (Lin et al., 1991). Above the following reason, SEM gave the addition explanation of data that the particle size was reduced. And the solubility test of IDM in the present of β-CD, was more soluble and increased when increasing the amount of β-CD. Kneaded mixtures of α-CD gave lower dissolution than the others. IDM-CDS mixtures because they had no interaction with IDM this evidenced of no interaction can be supported from IR spectra, DTA thermograms which were identical to those of pure drug. The α-CD kneaded mixtures possessed the high dissolution than pure drug and kneaded drug because the particle size reduction and reduce agglomeration of the IDM particles when incorporated with α-CD.

It is considered that a critical mixture ratio is present for dissolution of IDM from IDM kneaded mixtures and this ratio was approximately 1:2, 1:1, 1:0.5 in β-CD, δ-CD and DIMEB, respectively (Figures). From the above statement, when the weight ratio of CDS increased above this value, no enhancement was observed in drug release. In addition, the ratio above 1:3, β-CD gave nearly the same dissolution profile as δ-CD and DIMEB at 1:1 ratio. This result showed that the inclusion complex in β-CD kneaded mixture markly increase in the dissolution.

During the dissolution tests the powders of pure drug and kneaded drug were poor wetted because they floated on the medium

surface thus hindering dissolution. This poor wettability resulted in low dissolution of pure drug and kneaded drug. When the quantity of CD in kneaded mixtures increased, the more particles sank to the bottom of the flask or rapidly dissolved in the dissolution medium, which indicated that CD may reduce the surface tension of the medium and facilitate the wetting. These results leading to a higher dissolution especially β-CD has surfactant-like properties (Lin and Kao, 1989). In the case of DIMEB, the decrease in surface tension with increase in concentration had been reported (Hirayama et al., 1988), this leading to the high dissolution profile of both kneaded mixtures.

3. Solvent mixtures

In the coevaporated of DIMEB at the ratio 1:1, the high dissolution at the initial stage was observed and slightly increased after 25 minutes. The high dissolution at the first stage possibly came from polymorphic transformation the result was supported by DTA thermograms which indicated Form III presented in the products. In addition IR spectra gave carbonyl stretching shift of carboxylic group to the higher wave number (1717 to 1732 cm^{-1}) and carbonyl stretching of ketone group to the lower wave number (1692 to 1687 cm^{-1}) indicated some interaction of DIMEB and IDM in the mixtures. In the case of slowly increase in dissolution at later stage, this phenomena could be explained by SEM which revealed the small fine needle of Form II on the surface of the products, X-ray diffraction also exhibited a sharp peaks with corresponding to

treated drug. Moreover, during the examination of dissolution, it was found that the aggregated and coalesced of particles had been formed to a hard compact on the test solution and still remained until 60 minutes. The hard compact might be Form II polymorph which exhibited slow dissolution. This could be hypothesized that during the dissolution, Form III was rapidly soluble and partly change to Form II that made IDM particles to aggregate which exhibited slow dissolution. The similar result of polymorphic transition in the medium also was in agreement with the previous report (Corrigan, Holohan and Reilly, 1985). In the ratio 1:2 and 1:3, dissolution profile obtained the highest over other CDS. Because of the coevaporated was rapidly dissolved in dissolution medium and IR spectra showed a shift of carbonyl stretching of carboxylic group to the higher wave number (1717 to 1735-1736 cm^{-1}), in contrast carbonyl stretching of ketone group to the lower wave number (1692 to 1686-1687 cm^{-1}). This may be due to the interaction of IDM and DIMEB in the coevaporated mixtures. Furthermore, SEM obtained the smooth surface of coevaporated mixtures liked treated DIMEB and could not be seen the IDM particles, DTA exhibited no endothermic peaks and occurred the halo pattern on the X-ray diffraction. This absence of IDM diffraction may indicated that an amorphous form might be exist in the mixtures.

In α-, β- and δ-CD, The following of enhanced dissolution was ranked as follow : δ-CD > β-CD > α-CD. The increase in dissolution was due to the polymorph change in three products, this was the mixture of Form II and Form III because of in IR

spectra showed the typical spectra of polymorph Form II while DTA thermograms showed the endothermic peak of polymorph Form III. The superiority of ϑ-CD over β-CD can be explained that ϑ-CD was more soluble than β-CD hence the more solubilization effect resulted. However, α-CD was more soluble than β-CD, but the dissolution of β-CD was higher than α-CD. This possibly due to during the solvent method preparation, there were some or partly inclusion complexs occurred in β-CD solvent mixture while α-CD did not. This could be seen by DTA thermogram.

SEM photomicrographs

Examining the SEM photomicrographs, the plates and prism of IDM was reduced in grounded drug, agglomerate to coarse particle in kneaded drug and turned into bundle of aggregate of needle shape in treated drug. For grounded drug the dissolution was increased due to size reduction or improved in wettability; however, decreased dissolution in kneaded and treated drug were observed. These may owing to the large particle size and increase in agglomerate of IDM particles that would reduce the surface area in contact with dissolution medium.

In grounded mixtures and kneaded mixtures, the size of IDM was reduced and some interaction that may be inclusion complex occurred during the preparation in β-CD, ϑ-CD and DIMEB. Furthermore, in the case of DIMEB , polymorphic transformation was also observed as indicated from DTA. As was mentioned in dissolution studies, the

changed of IDM in α-CD mixtures was not found. These would make β-CD, δ-CD and DIMEB obtained the high dissolution over α-CD.

In coevaporated DIMEB mixtures. IDM particles disappeared and the products obtained was similar as treated DIMEB and yellow color appeared. In the case of ratio 1:1, the small fine needle in the products was also observed. This may be due to the polymorphic change as explained in dissolution studies. In α-, β- and δ-CD solvent mixtures,there were size reduction and changing in crystal habit in the products. Moreover, there were some interaction between IDM with β- and δ-CD that may be inclusion complex during the preparation could cause the high dissolution obtained.

IR spectra

The dispersed state of the IDM molecules in various CDS were investigated and analysis by the infrared spectra especially in the carbonyl stretching regions. When the drug was treated with alcohol, the IR spectra showed peaked at 1650, 1692 and 1753 cm^{-1} indicated the transformation of polymorph I to polymorph II. In grounded mixtures and kneaded mixtures of four CDS, a simple superimposition of the pure drug and CDS spectra could be noticed; moreover, the intensity of the C-O stretching band was reduced when the quantity of CD increased. This lower intensity of the carbonyl stretching band at 1690 and 1717 cm^{-1} may due to the effect of dilution of the drug by means of CDS or the occurrence of inclusion compound in β-CD and δ-CD. This could not be seen clearly because

the inclusion compound occurred only partially. In the coevaporates of DIMEB, there was a significant shift in carbonyl band indicated some interaction between IDM and DIMEB. In the case of α-, β- and δ-CD prepared by solvent deposition, the carbonyl stretching band was similar to treated drug that indicated that IDM had polymorphic changed from IDM polymorph Form I to Form II.

DTA thermograms

DTA thermograms of pure drug exhibited the endothermic peak at 160 °C, indicated the polymorph Form I. In grounded drug and kneaded drug the same polymorph Form also obtained because there were no shift of endothermic peaks. Treated drug showed two endothermic peaks, one sharp at 153 °C indicated the transformation of polymorph I to II and the other broad peak at 310 °C could referred to the decomposition of IDM.

Cyclodextrins have no defined peak for melting point, but from above 200 ° C they begin to decompose. The observed thermoanalytical peak properties of CDS depend on the water content, crystal structure, heating rate and atmospheric composition. DTA thermograms of the anhydrous α-CD show exothermic peaks at 167 °C and small endothermic peaks at 230 °C. When α-CD was not completely dry, three endothermic peaks are present, presumably reflecting absorbed and bounded water of different energies. These peaks were centered at 80 ° C, 106 °C and 129 °C (Szejtli, 1988). In this experiment, α-CD showed two broad endothermic peaks around 50 °- 60 °and 78 °- 88 °C and

small endothermic peak at 137°- 138°C, these may indicated the absorbed and bounded water escape from α-CD molecules. Afterthat, there was no endothermic peak until around 310°C, one large broad and irregular endotherm could be seen to refer decompose of α-CD. β-CD and δ-CD exhibited two endothermic peaks, one broad peak around 60° - 110° C referred the water escape and the other broad and irregular endotherm after 310°C indicated the decomposition of β-CD and δ-CD. However, DIMEB did not showed endothermic peak around 100° - 300° C.

In grounded mixture and kneaded mixture of α-,β- and δ-CD, the thermograms was the combined features of thermograms of each component also indicated no interaction. However, the IDM diffraction peaks were decreased in the intensity while CD peaks increased with increasing the amount of CD. This indicated the IDM particles were dispersed as individual crystals in the mixtures that made the improvement in wettability. In the case of β-CD, a small endothermic peak at 215°- 217° C occurred in both grounded and kneaded mixtures eventhrough at small ratio. And this endothermic peak increased with increasing the amount of CD while IDM peak decreased.The occurrence of the new peak indicated some interactions between IDM and β-CD and created the new compound that may be inclusion complex. Because of IDM endotherm did not disappear so the product was the mixture of IDM drug dispersed in β-CD and inclusion compound.

Grounded mixtures, kneaded mixtures and 1:1 coevaporated of DIMEB, there were a shift of IDM thermogram from 160°to 147°- 150°C indicated the polymorphic changed to Form III. But in the 1:2 and 1:3 coevaporated systems, no endothermic peak of IDM were found. It displayed some interaction between DIMEB and IDM. This may be referred to an amorphous form of IDM in DIMEB. In solvent deposition of α-, β- and δ-CD, the peaks at 147°- 150°C were observed. This referred the polymorphic transition to Form III in the mixtures. Furthermore, in the case of β-CD solvent mixtures, partially complex also occurred because the endothermic peak at 217 °C could be seen.

X-ray diffraction studied

Treated drug with alcohol showed the typical X-ray diffraction peaks indicated that polymorph I in pure drug was changed to Form II. From X-ray diffraction in grounded and kneaded mixture of DIMEB and IDM, indicated change in diffractogram at 5.4°,6.9°,7.8°,8.6°,11.0°,11.5°,16.9° and 20.8°.The results can be supported by DTA thermograms which may be indicated polymorphic transition to polymorph III and existed in this mixtures. In the case of 1:1 coevaporated, some diffraction peak of IDM were also obtained. These indicated the partly crystalline IDM still remain in the coevaporates. However, in 1:2 and 1:3 coevaporated, the absence of crystalline IDM peaks were observed. This might be indicated that IDM present as an amorphous form in DIMEB coevaporate systems.

Phase solubility studies

It was observed that the concentration of IDM increased with increasing of the concentration of β-CD and DIMEB. In the case of α- and δ-CD slightly increase in the solubility of IDM were found. The phase solubility diagrams of β-CD and DIMEB can be classified as type A_N which indicated the formation of inclusion compound in the solution. These results were the same as previous reported (Müller and Brauns, 1985).

The summary of dispersion methods used to prepare IDM-CDS dispersed systems might increase dissolution which could be explained as follows:

1. Size reduction, deaggregation or deagglomeration of particles (SEM), hence the specific surface area of drug particles might increase and so did the dissolution rate.

2. A partially inclusion complex formation occurred during the preparation or in the dissolution medium (DTA, IR, X-ray diffraction, SEM and phase solubility studies).

3. The possible solubilization effect by CDS in the microenvironment (diffusion layer) immediately surrounded the drug particle.

4. Polymorphic change of drug molecule from the low solubility polymorph to the high ones, or in amorphous form. This can caused a significantly higher solubility than the crystalline phases. However, in some cases the high solubility polymorph which is unstable, being converted to crystalline Form I and II thus retard the dissolution.

5. Wetting characteristic and dispersibility of drug powders in the CDS were improved. This may be due to the surfactant-like property of CDS hence reduced surface tension of the medium thus increasing in dissolution (Hirayama et al., 1988; Müller, Brauns and Backensfeld, 1988; Szente and Szejtli, 1987).

It is not always necessary to prepare true inclusion complexes to improve dissolution of IDM. Because of physical mixtures of IDM and CDS in this study show faster dissolution rates than drug alone. Furthermore, the preparation of the true inclusion complex is rather difficult and mostly impractical for preparation of commercial product.

From the dissolution profile and T80%, DIMEB showed the highest dissolution followed by ǒ-CD, β-CD and α-CD, respectively. Dispersion methods that obtained in the highest dissolution were

ranked as follow: coevaporate > kneading > grinding for DIMEB and kneading > grinding > solvent deposition for α-, β- and δ-CD. These results of the studies indicate that CD could be useful to increase dissolution profile of IDM. The physicochemical property of IDM in dispersed systems could be used to explain why such dispersed products gave the high dissolution or predict the outcome of the final product obtained. Dispersion method, type and quantity of CDS used also influenced the solubility of drug. Furthermore, the cost of CDS especially β-CD derivatives were high then these should be considered in order to get the ideal desirable properties of CDS may be succeeded in enhancement of dissolution of IDM.

Capsule evaluation

Because of DIMEB was the most powerful CD to increase dissolution of IDM by various methods of preparations in this study. Each preparation methods of DIMEB and IDM mixtures at the lowest ratio that could pass the USPXXII requirements (80% IDM dissolved within 20 minutes) was selected and subjected to prepare capsule for comparison the dissolution with corresponding IDM powders.

The increasing in dissolution of seven IDM capsules prepared were ranked as follow : 1:2 coevaporation > 1:0.5 kneaded mixture > 1:0.5 grounded mixture > grounded drug > treated drug> pure durg > kneaded drug. These reasons were the same as previous discussion in powders dissolution. The slow dissolution at the initial stage was due to the lag time of the capsule to dissolve. In pure drug,

grounded drug, treated drug and kneaded drug capsule, the dissolution were higher than that of powder because of the possible solubilizing effect of lactose in the preparation. Lactose was used as diluent and it was very soluble in the medium hence enhanceing the dissolution. However, the dissolution of grounded mixture, kneaded mixture and coevaporated capsule were lower than that of the corresponding powders. In grounded mixture, kneaded mixutre and coevaporated powders, they were observed that IDM in those three systems appeared to be high polymorph or amorphous Form. When incorporated with diluents such as lactose, corn starch, stearic acid in the capsule formulation, the polymorphic change may be occurred by the influence of these diluents that caused the low dissolution. However, the lower of IDM capsules, T80% of three mixtures capsules passed the requirement of the USPXXII specifications.

It would be stated that although the dispersed mixtures offered increasing in dissolution, the particle size of the products might be effect on the dissolution. Therefore, the optimum particle size should be explored. Furthermore, further investigation must also be studied as follows:

1. Stability of the coevaporated products in powder or when included with other excipients should be studied including dissolution and physicochemical properties.

2. IN-VIVO test; to determined the bioavailability. General, higher blood levels can be achieved if poorly soluble drugs are solubilized by CD but not always. If the high blood level is not desirable, the dose of drug could be adjusted.

3. Manufacturing process and the unit operation should be operated appropriately in order to find the optimization point that obtained the desirable dissolution profile of drug.

CONCLUSION

Dispersed systems of IDM with CDS yielded more rapid dissolution than pure drug and corresponding powders. The CDS gave faster disolution characteristics of indomethacin were ranked as follow : DIMEB > δ-CD > β-CD > α-CD. Dispersion techniques used, type, quantity of CDS and physicochemical properties of IDM in the mixtures also influenced in the dissolution profiles obtained. From X-ray diffraction, IR spectra, DTA studied and SEM, an important role for improving the dissolution characteristics of IDM in dispersed systems were the presence of high soluble polymorph of IDM or amorphous Form and partially inclusion complex occurred in the preparation and / or test solution. Furthermore, the improvement in the wettability, the solubilization of the drug by CDS and size reduction may also contribute to the enhancement of dissolution of IDM dispersed in CDS.

The polymorphic conversion from high energy polymorph to the low ones might occur when the dispersed systems were incorporated with diluents used in the capsule formulation. These caused the lower dissolution of the prepared capsule obtained.