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INTERACTION OF ROFECOXIB WITH β-CYCLO-DEXTRIN AND HP-β-CYCLODEXTRIN IN AQUOUES SOLUTION AND IN SOLID STATE

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تم فى هذا البحث دراسة التفاعل بين عقار الروفيكوكسيب المضاد للالتهابات والمسكن للألم ، مع كل من بيتا سيكلودكسترين والهيدروكسى بروبيل بيتا سيكلودكسترين فى الحالة السائلة والحالة الصلبة. وقد تم تعيين ثابت التكوين فى درجات حرارة مختلفة باستخدام طريقة منحنى الاذابة. تم تحضير انظمة ثنائية للعقار مع السيكلودكستريات المستخدمة باستخدام طريقتى الطحن المتزامن والترسيب المتزامن. وقد تم دراسة مدى تكون متراكبات متداخلة للعقار مع السيكلودكستريات المستخدمة بواسطة التحليل السعرى التفاضلى ، والأشعة تحت الحمراء وأشعة إكس. تم دراسة معدل اتاحة العقار من الأنظمة الثنائية المحضرة ومقارنتها بكل من المخلوط الفيزيائى للعقار مع السيكلودكستريات المستخدمة والعقار بمفرده. وقد أظهرت الدراسة زيادة معدل ذوبان العقار فى المحاليل المائية معتمدا على كل من تركيز السيكلودكسترين ودرجة الحرارة. ولقد تم تحديد منحنى الاذابة ووجد انه يتبع نوع A_L والذى يعنى أن عقار الروفيكوكسيب يكون متراكبا متداخلا بنسبة ١:١ مع كل من بيتا سيكلودكسترين والهيدروكسى بروبيل بيتا سيكلودكسترين. ولقد وجد أن ثابت التكوين يساوى ١٠٤,٤٥ مول$^{-1}$ و ١٢١,٦٥ مول$^{-1}$ على التوالى. وقد أظهرت طريقة الطحن المتزامن زيادة معدل الاتاحة للعقار بالمقارنة بالطرق الأخرى. وتم ترتيب الزيادة فى معدل اتاحة الروفيكوكسيب من النظم المحضرة مع السيكلودكستريات محل الدراسة كالتالى:المخلوط المحضر بطريقة الطحن المتزامن ثم المخلوط المحضر بطريقة الترسيب المتزامن ثم المخلوط الفيزيائى وأخيرا العقار بمفرده. وقد وجد أن فترة نصف عمر العقار المحضر بطريقة الطحن المتزامن مع كل من الهيدروكسى بروبيل بيتا سيكلودكسترين وبيتا سيكلودكسترين ٧ و٥٠ دقيقة على التوالى.

*The interaction between rofecoxib (ROF), an analgesic anti-in*flammatory drug, with β-*cyclodextrin and HP*-β-*cyclodextrin was evaluated in aqueous environment and in solid state. The solubility* of ROF with β-*CyD and HP*-β-*CyD in aqueous solution was*

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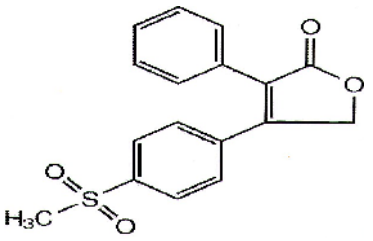
determined and the stability constants were calculated from the phase solubility studies at different temperatures. Binary systems of ROF with the investigated CyDs were prepared by co-grinding and solvent evaporation methods. The formation of inclusion complexes with β-*CyD and HP*-β-*CyD in the solid state was investigated by differential scanning calorimetery, infrared spectroscopy and X-ray diffractometry. Dissolution rate of ROF binary systems was determined and compared with those of the physical mixture and the pure drug. It was found that the solubility of ROF increased as a function of both CyDs concentration and temperature showing an* A_L*- type diagram indicating the formation of 1: 1 stoichiometric inclusion complexes. The apparent association constants were found to be 104.45* M^{-1} *and 121.65* M^{-1} *for* β-*CyD and HP*-β-*CyD; respectively. Co-grinding method led to enhancement of ROF dissolution rate in comparison to the other preparation methods. The in vitro dissolution rate of ROF at pH 7.4 could be ranked in the following order: ground mixture, coevaporate, physical mixture and pure drug. Ground mixture of ROF with HP-* β-CyD and β-*CyD has a* $t_{50\%}$ *= 7 min and 50 min, respectively.*

INTRODUCTION

Cyclodextrins (CyDs) are torus – shaped oligosaccharides consisting of 6, 7 or 8 (α – β – and γ CyDs; respectively) glucopyranose units through α – 1.4 – linkages with hydrophobic central cavity and hydrophilic exterior surface. The entire or at least partial inclusion process of some drugs into CyDs has led to improvement in a variety of physicochemical and pharmaceutical properties such as aqueous solubility,[1&2] dissolution rate,[3&4] local irritation[5] and bioavailability of many drug molecules.[6] In addition, inclusion of molecules within the cavity of CyDs may protect the guest molecules from the external environment, and hence, CyDs may be used to optimize the chemical stability of molecules susceptible to degradation.[7&8]

Inclusion complexes of nicardipine hydrochloride, a calcium-channel antagonist, with β-CyD or HP-β-CyD, were prepared using different techniques (kneading, evaporation, freeze-drying and spray-draying). It was found that all the combinations with HP-β-CyD were more effective in achieving the enhancement of the nicardipine dissolution rate, yielding better performance than the corresponding ones with β-CyD.[9] The effect of HP-β-CyD on the aqueous solubility and chemical stability of CKD-732, a new angiogenesis inhibitor, was investigated with an aim of preparing a stable and effective parenteral

formulation. The results demonstrated that the CKD-732 - HP-β-CyD complex is an attractive formulation for use in the parenteral delivery of CKD-732.[10]



ROF is described chemically as 4-[4-(Methylsulfonyl)phenyl]-3-phenyl-2(5H)-furanone derivative. It is a selective cycloxygenase-2-inhibitor administered orally as an analgesic anti-inflammatory drug and it is structurally and pharmacologically related to celecoxib[11&12] with less ulcerogenic effect[13-15] so, it is a good candidate for oral dosage forms. ROF is practically insoluble in water and as such, its oral absorption is dissolution rate limited. ROF is available in the market in the form of tablets and oral suspension only.[11] Unfortunately, the release of ROF from the marketed tablet is limited because of the very poor aqueous solubility of the drug a factor that led to variable bio-availability.[16] However, by increasing the solubility and dissolution rate of the drug better release can be obtained. In a previous work,[16] kneaded and physical mixtures of ROF with β-CyD were prepared and investigated by DSC, X-ray and dissolution studies. It was found that the dissolution rate of the drug was improved by complexation. However; this method for complexation (kneading) is not a commercial method.

The objective of this study is to prepare ROF – CyDs complexes by milling method which could be more economic[17] and by coevaporation technique. The prepared solid systems were characterized using DSC, IR, X-ray diffraction patterns as well as dissolution. Solubility studies were also performed.

EXPERIMENTAL

Materials

ROF (Rofecoxib)kindly supplied by Egyptian International Pharmaceutical Industries Co., (E.I.P.I.Co.), Egypt, β-cyclodextrin was purchased from Sigma, Chem. CO., U.S.A., 2-Hydroxypropyl-β-cyclodextrin (D.S. value-RMN, 0.840) was purchased from Pharmatec Inc. Alachua, Florida, USA. All other chemical reagents were of analytical grade and used as received.

Equipment

Thermostatically controlled temperature water bath (KARL KOLB-Germany), vibrational uniball mill (VEB leuchtenbau-KM1, Germany), Shimadzu - 470 Infrared Spectrophotometer (Japan), Shimadzu DSC-50 (Japan), X-Ray diffractometer Philips 1710 diffractometer (Germany), SR6 Dissolution Test Station, Hanson Research Corporation (California, USA).

Methods

1- Solubility study

Solubility studies were performed as described by Higuchi and Connors.[18] An excess amount of ROF (about 10 mg) was added to 50 ml volumetric glass flasks. Cyclodextrin solutions of different concentrations (0–10 mM of β-CyD or HP-β-cyclodextrin) were prepared in distilled water. A constant volume (10 ml) of the investigated cyclodextrin solutions was added to each flask. The flasks were closed and brought to solubility equilibrium at 25º and 37º ± 0.5° after shaking over a period of 24 hours.[16]

After equilibrium was reached (24 hours), the contents of each flask were filtered through a disk filter 0.45 µm (Millipore filter). The filtered solutions were appropriately diluted and the amount of ROF solubilized was determined spectrophotometrically at 264 nm against a suitable blank similarly treated. The drug content was determined from the linear regression analysis based on the standard calibration curve.

2-Preparation of the physical mixture and inclusion complexes

Preparation of the physical mixtures

Physical mixtures of ROF-β-CyD and ROF-HP-β-CyD in a molar ratio of 1:1 were prepared by simple blending in a glass mortar for five minutes.

Preparation of inclusion complexes

Co-evaporation method

Co-evaporates of ROF-β-CyD and ROF-HP-β-CyD were prepared by solvent evaporation method at a molar ratio of 1:1. ROF was dissolved in a sufficient volume of acetone. β-CyD and HP-β-CyD were dissolved in distilled water at 40º. The drug and the respective CyD solutions were mixed together with constant stirring for about one hour. The solutions were evaporated at 40º under vacuum until a constant weight. The co-evaporate was sieved to obtain a particle size range of 125-250 µm then stored in a dessicator over calcium chloride until analyzed spectrophotometry for their drug content.

Co-grinding method

The ground mixtures of ROF with either β-CyD or HP-β-CyD in a 1:1 molar ratio were prepared by co-grinding method using the vibrational uniball mill for about 15 minutes (an optimum time which was obtained by experiment). The ground mixtures were sieved to obtain a particle size range of 125-250 µm then stored in a dessicator until analysis.

3-Characterization of the prepared systems

Differential scanning calorimetry studies

DSC curves were obtained by using a Shimadzu DSC-50 equipped with a software computer program. Samples of about 5 mg were placed in an aluminum pan of 50 µl capacity &

0.1 mm thickness press-sealed with aluminum cover of 0.1 mm thickness. An empty pan sealed in the same way was used as a reference. Thermograms were measured by heating the sample from 30 - 250º at a rate of 10º min^{-1}, under nitrogen flow of 40 ml/min. Indium was used as standard for calibrating the temperature. Reproducibility was checked by running the sample in triplicate, the standard deviations calculated were found negligible.

X-ray diffraction studies

The X-ray diffraction patterns for the selected samples with a particle size range of 125-250 µm were determined using a computer Philips 1710 operating in two modes using CuKα radiation. A Cu target tube operated at a voltage of 40 kV and a current of 40 mA and a single crystal graphite monochromator were employed. A scanning speed of 0.6°/min and a wide angel diffraction of $4°<2\theta<60°$ was employed. An attached microprocessor utilizes a special software program to analyze peak position and intensities. Standard polycrystalline silicon powder was used to calibrate the instrument.

Infrared analysis (IR)

IR analysis was performed for ROF, β-CyD, HP-β-CyD and the prepared systems using Shimadzu - 470 Infrared Spectrophotometer. Samples of 1-2 mg were mixed with potassium bromide (IR grade). The obtained mixture was then compressed into discs in a compressor unit under vacuum then scanned from 4000 to 600 cm^{-1} with an empty pellet holder as a reference.

Determination of drug content of the prepared samples

An accurately weighed sample of the prepared systems equivalent to 12.5 mg of the drug was introduced into a 100 ml volumetric flask, then dissolved in a minimum amount of acetonitirle and completed to 100 ml with phosphate buffer of pH 7.4. After suitable dilution, ROF content was determined spectrophotometrically at 264 nm. Only those samples containing 100 ± 5% of the claimed amount of ROF (12.5 mg) was considered for further studies.

In-Vitro dissolution studies

Dissolution experiments were carried out in triplicate with USP dissolution apparatus type II at a rotation speed of 100 rpm. Samples of 12.5 mg of ROF or its equivalent of the physical mixtures, coevaporates and ground mixtures with either β-CyD or HP-β-CyD were placed into 900 ml of the dissolution medium. (Phosphate buffer of pH 7.4, kept at 37±0.5º). At appropriate time intervals, 5 ml of aliquots were withdrawn and filtered with disc filter pore size 0.45 µm. Equal volumes of buffer kept at 37º ± 0.5º, were added as displacement to the withdrawn samples. The samples were analyzed spectrophotometrically at 264 nm. It was found that none of the additives used interfered with the spectrophotometric assay of the drug. The

mean of three determinations was considered.

RESULTS AND DISCUSSION

Solubility studies

Figs. 1 and 2 and Table 1 show the effect of different concentrations of β-CyD and HP-β-CyD on the aqueous solubility of ROF at 25º and 37º; respectively. It was found that the solubilizing effect of HP-β-CyD for the drug was higher than that of β-CyD. This may be due to the low aqueous solubility of β-CyD (about 1.8% w/v, at 25º) in comparison to hydroxypropyl-β-cyclodextrin whose solubility in water is about (50% w/v) at 25º.[19] The aqueous solubility of ROF was linearly increased as a function of β-CyD or HP-β-CyD concentration and shows features of an A_L type phase solubility diagram.[18] These results suggested that water – soluble 1:1 ROF cyclodextrin complex was formed. Similarly, it was found that HP-β-CyD gave a higher solubilizing efficiency for nifedipine than that of β-CyD.[20]

Stability constants were calculated assuming a 1:1 stiochiometry. The stability constant of the formed complex was calculated from the slope of the linear line using the following equation.[18]

$$K = S / (1 – S) S_{\circ}$$

Where K= stability constant, S= slope and $S_{\circ}$ = solubility of the drug in water. ROF / HP-β-CyD complex has a higher value of K (apparent stability constant) than that of ROF / β-CyD complex. The apparent 1: 1 stability constants (K) of the ROF / β-CyD and ROF / HP-β-CyD were found to be 73.75 M^{-1} and 101.73 M^{-1}; at 25° respectively. The effect of temperature on the aqueous solubility of ROF in β-CyD and HP-β-CyD is shown in Figs. 1 and 2 and Table 1. There is an increase in the solubility of ROF upon increasing the temperature. It was noticed that there is an increase in the stability constant with increasing the temperature, which means that the interaction of the drug with cyclodextrins at higher temperature was stronger.

The apparent 1:1 stability constants (K) of ROF / β-CyD and ROF / HP-β-CyD were found to be 104.45 M^{-1} and 121.65 M^{-1}; respectively at 37º. These results were consistent with the increasing water solubility of the drug by raising the temperature as shown in Table 1. The increase in the solubility of ROF may be attributed to the positive temperature effect on the free drug and complexed drug solubility. Also, this may be related to the liberation of water molecules bound in the cavity of CyDs which were substituted by the guest molecules. Venetura *et al.*[21] studied the inclusion complexation between 2-biphenyl acetic acid and different CyDs in aqueous environment. They found that increasing the temperature from 25 to 37 and 45º led to enhancement of the intercept value for all complexes and this behavior could be due to the positive effect of the temperature on the complex solubility.

Table 1: Effect of different concentrations of β-CyD or HP-β-CyD on the aqueous solubility of ROF at 25 and 37º.

Concentration of ß-CyD or HP- ß-CyD (mM)	Amount of ROF solubilized (mM x 10^2)			
	ß-CyD		HP- ß-CyD	
	25º	37º	25º	37º
0	6.85	8.16	6.85	8.16
2	8.03	9.43	8.57	10.10
4	9.16	11.10	10.06	12.11
6	10.16	13.08	11.54	14.01
8	11.19	14.5	12.69	15.78
10	11.93	16.26	14.04	18.10

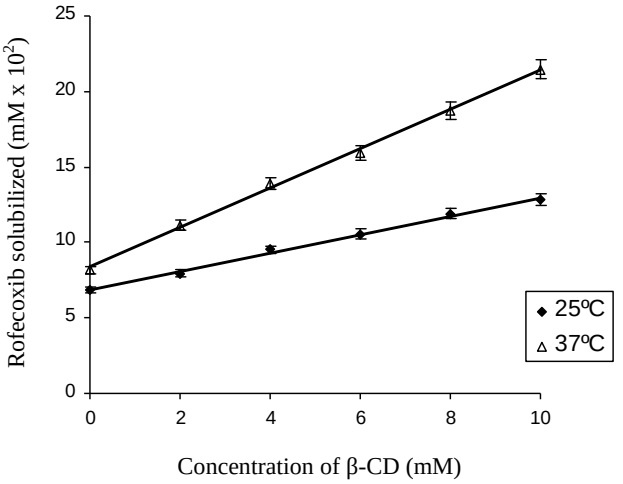


Fig. 1: Phase solubility diagrams of ROF in β-cyclodextrin aqueous solutions at 25 and 37º.

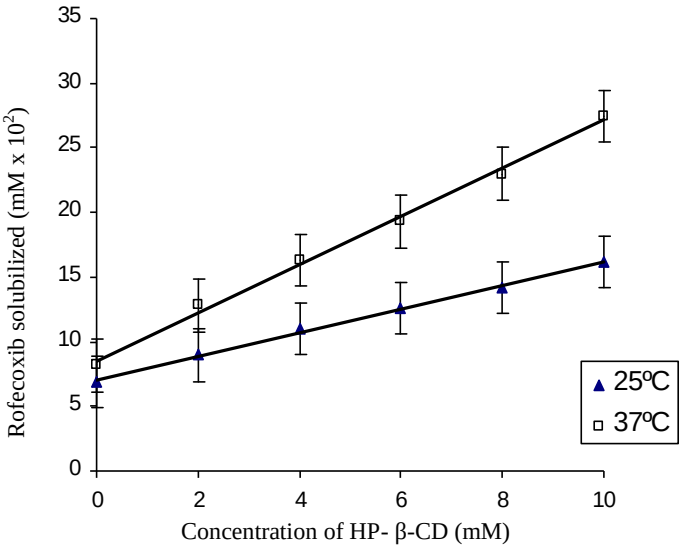


Fig. 2: Phase solubility diagrams of ROF in HP-β-cyclodextrin aqueous solutions at 25 and 37º.

Differential scanning calorimetry studies

Fig. (3) shows the DSC thermograms of pure ROF, β-CyD, HP-β-CyD, its corresponding physical mixtures and coevaporates as well as ground mixtures in 1:1 molar ratio. ROF shows an endothermic peak with an onset temperature of 209.5º, which corresponds to its melting point. The peaks corresponding to the evaporation of water vapors from β-CyD and HP-β-CyD appeared at 80º[22&23] and 60º, respectively, (Fig. 3 traces D, H). It is evident that the thermograms of physical mixtures of both the drug and either β-CyD or HP-β-CyD are superimpossible. The physical mixtures and coevaporates of ROF: β-CyD show an endothermic peak at 209.5º, due to fusion of the drug (Fig. 3 traces, A, B respectively). The drug melting peak at 209.5º shifted to 207.8º in ground mixture of ROF: β-CyD. The appearance of endothermic peak near 207º for the ground mixture of the drug with β-CyD could still reflect the presence of a few drug crystals in the ground mixture, but the area of this endotherm (ΔH = -15.21 J/g) is reduced in comparison with pure ROF crystals (ΔH = -91.5 J/g) This can be attributed partially to the dilution effect when this ratio was prepared. Also, the melting peak of β-CyD in all the above prepared systems was shifted from 80º to 60º. This result could be due to incomplete complex formation when low ratio of β-CyD was used (1:1 molar ratio). These results are in accordance with Franciso *et al.*,[24] who suggested that the physical mixtures as well as coevaporate mixtures of tolbutamide with β-CyD showed incomplete complexation and

the endothermic peak characteristic to the drug was still present in these prepared systems.

In case of coevaporates and ground mixtures of ROF: HP-β-CyD 1:1 molar ratio it was observed from Fig. (3) that the water evaporation peak of HP-β-CyD at 60º slightly disappeared in case of ground mixtures which indicates that the drug had penetrated into the HP-β-CyD cavity replacing the water molecules. The drug melting peak at 209.5º shifted to lower temperature at 208.4º in case of physical mixtures (Trace E), at 205.9º in coevaporates (Trace F) and at 198º in ground mixtures (Trace G) coupled with a decrease in their intensities. The peaks at 208.4 and 205.9º can still reflect the presence of a few drug crystals in the physical mixture and coevaporate preparations, respectively; however, in case of ground mixture the thermal effect appeared more broadened and highly reduced in intensity, which suggest some drug – cyclodextrin interaction.

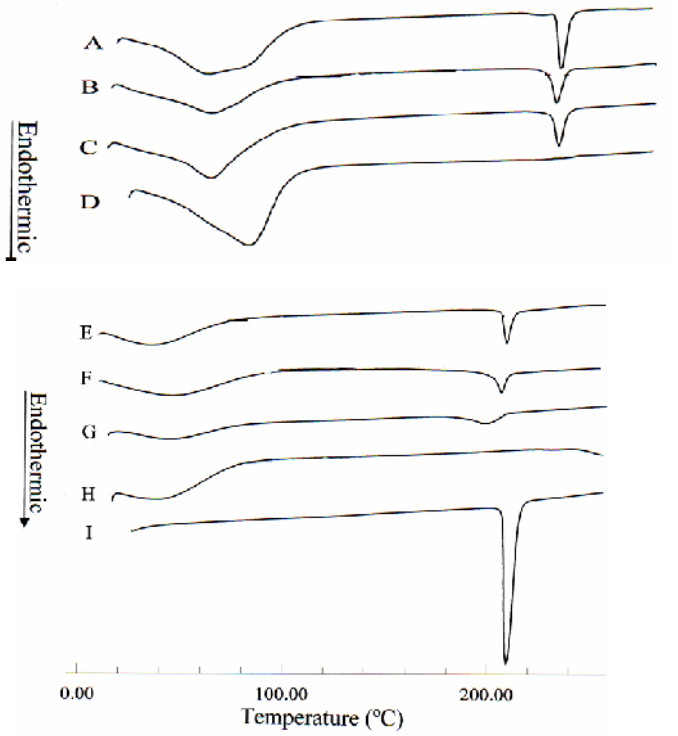


Fig. 3: DSC thermograms of ROF / β-cyclodextrin and HP-β-cyclodextrin systems in (1:1 Molar ratio).

(A) Physical mixture with β-CyD, (B) Coevapoarte with β-CyD, (C) Ground mixture with β-CyD (D) β-CyD alone, (E) Physical mixture with HP-β-CyD, (F) Coevapoarte with HP-β-CyD, (G) Ground mixture with HP-β-CyD, (H) HP-β-CyD alone, (I) Drug alone.

X-ray diffraction studies

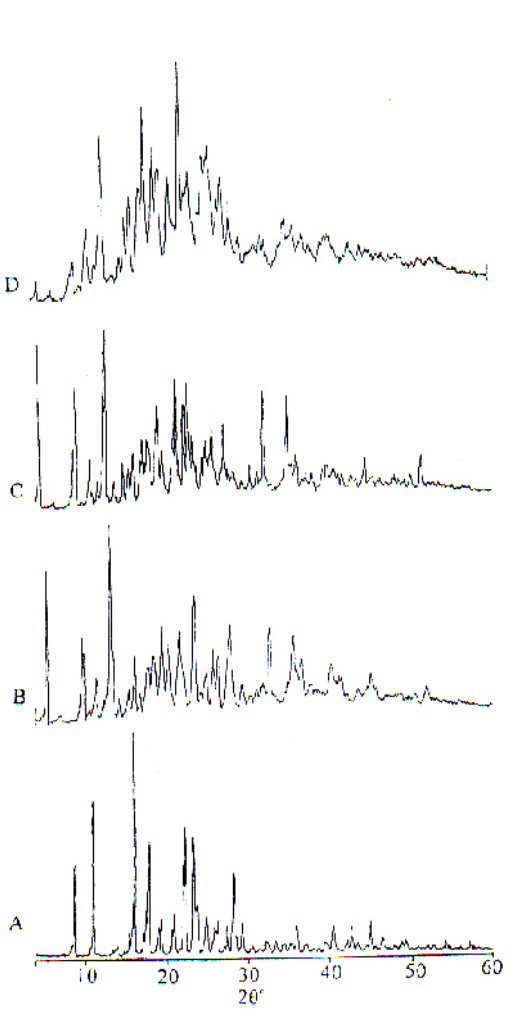
Powder X-ray diffractometry is a useful method for the detection of cyclodextrin complexation in a powder or microcrystalline states.[22] The X-ray diffraction patterns of pure ROF, β-CyD alone, their physical and ground mixtures in a 1:1 molar ratio are represented in Fig. (4). The diffraction patterns of ROF show important sharp intense diffraction peaks at (2θ) = 8.82˚, 11.08˚, 16.18˚, 17.94˚, 22.41˚ and 2θ = 23.39˚ (Figs. 4 and 5 trace A). The diffractograms of ROF and β-CyD exhibited a series of intense peaks, which are indicative of their crystallinity. The diffractogram of the physical mixture as well as the ground mixture were constituted practically by simple superposition of each component, indicating the presence of ROF in the crystalline state and inability to fit in the β-CyD cavity. Thus, it can be deduced that an equimolar of β-CyD is insufficient to complex all the drug using co-grinding method, these results are in agreement with DSC study.

Fig. (5) shows the powder x-ray diffraction patterns of the physical and ground mixture of ROF with HP-β-CyD in a 1:1 molar ratio. Grinding of ROF with HP-β-CyD shows appearance of new peaks at (2θ) = 16.9˚, 20.3˚ and 24.2˚ respectively, which is not present in the diffractograms of pure ROF or its physical mixture with HP-β-CyD. Also, it was clearly observed the disappearance of the crystalline peaks of ROF situated at (2θ) =17.9˚ and 22.4˚. These results may be attributed to an interaction between ROF and HP-β-CyD in the ground mixture suggesting the presence of a new solid structure with lower crystallinity than the drug where a possible inclusion of ROF inside the cyclodextrin cavity can occur. Similar observation also obtained from Sanghavi *et al*.,[25] who found that the X-ray diffraction pattern of the terfenadine inclusion complex was found to be diffused and different, confirming that, a new less crystalline solid phase was formed as compared to the physical mixture and this can be attributed to complex formation.

Infrared spectroscopy studies

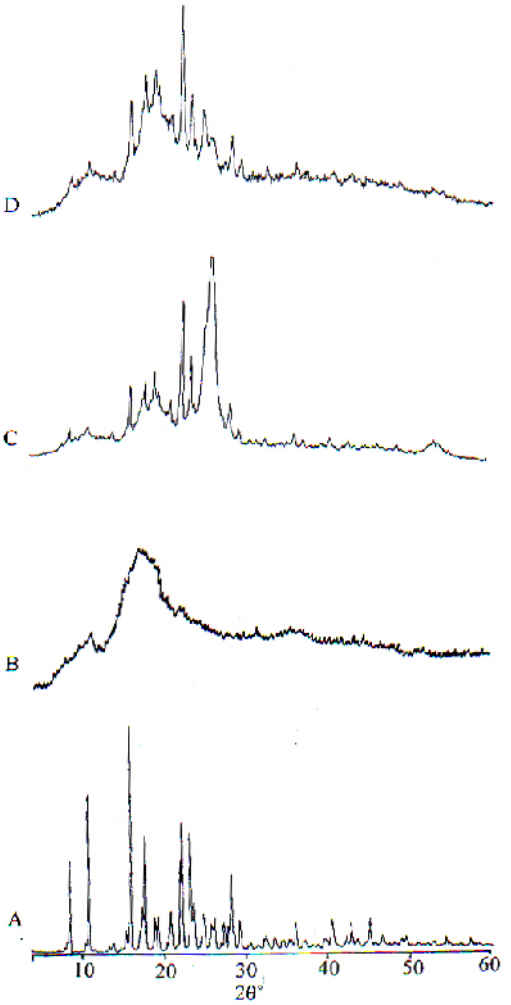
Figs. 6 and 7 show the IR spectra of ROF-β-CyD and ROF-HP-β-CyD systems. Infrared spectrum of ROF shows a peaks at 1743 cm^{-1}, which corresponds to (C=O) stretching of the carbonyl group of furanon ring at 1643 cm^{-1} for SO_2 group stretching and at 1303 and 1160 cm^{-1} for SO_2 bending. The IR spectra of β-CyD and HP-β-CyD, show absorption bands between 3300–3700 cm^{-1} for free hydroxyl groups vibrations and at 2970 cm^{-1} for bounded hydroxyl stretching, a broad band appeared at 1634 cm^{-1}, due to adsorbed water as seen in Figs. 6 and 7 trace B.

The decrease in the intensity of some bands i.e., carbonyl group (C=O) of furanone ring of the drug at 1743 cm^{-1} suggesting that a weak interaction might occur between ROF with CyDs during the preparation of the KBr disks[21] and due to dilution effect with the used CyDs.



ROF/β-CD

Fig. 4



ROF/HP-β-CD

Fig. 5

Fig. 4&5: X-ray powder diffraction patterns of different systems containing ROF / β-CyD and ROF / HP-β-CyD in 1:1 molar ratio.
(A) Drug alone. (B) β-CyD alone or HP-β-CyD alone, (C) Physical mixture, (D) Ground mixture.

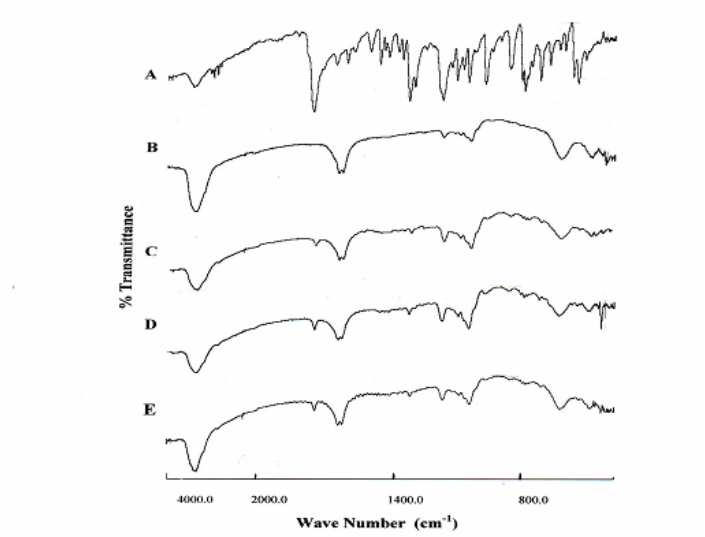


Fig. 6: IR spectra of ROF / β-cyclodextrin systems.
(A) Drug alone, (B) β-Cyclodextrin alone, (C) 1:1 M Physical mixture, (D) 1:1 M Coevapoarte, (E) 1:1 M Ground mixture.

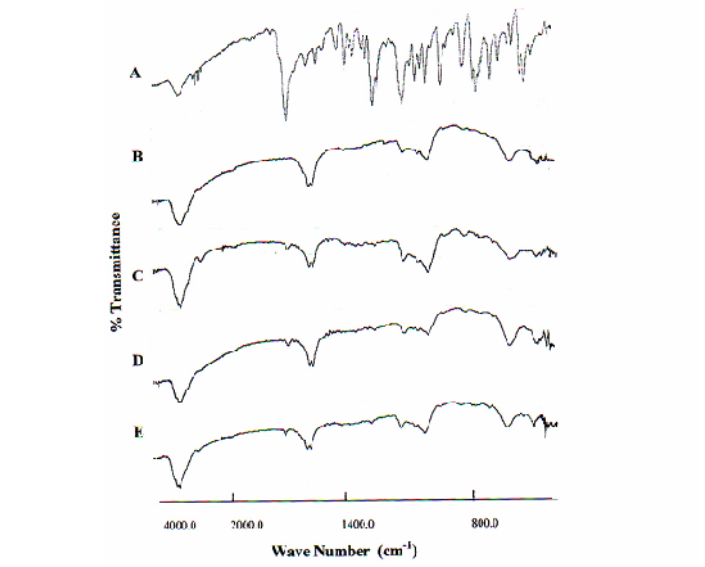


Fig. 7: IR spectra of ROF /HP-β-cyclodextrin systems.
(A) Drug alone, (B) HP-β-Cyclodextrin alone, (C) 1:1 M Physical mixture, (D) 1:1 M Coevapoarte, (E) 1:1 M Ground mixture.

The signals at 1303 cm^{-1} and 3150 cm^{-1} assigned to the symmetric bending vibration of SO_2 and $–CH_3$ functions of ROF respectively, were disappeared in ROF-HP-β-CyD ground systems which suggested these functional groups were included within the cyclodextrin cavity.[26] The broadening of the peak appeared from 3300 cm^{-1} to 3700 cm^{-1} which corresponding to free hydroxyl group of the CyDs, gave an indication about interaction of the drug with HP-β-CyD.

Dissolution studies

Fig. (8) shows the dissolution profiles of ROF powder, β-CyD, their corresponding physical mixture, coevaporate and ground mixture at molar ratio (1:1) in phosphate buffer at pH 7.4. The dissolution of the physical mixture of ROF with β-CyD in a1:1 molar ratio is slightly better than that of the untreated drug which dissolves less than 10% after 90 min. The coevaporate of ROF-β-CyD displays a dissolution rate greater than the physical mixtures but less than that of the ground mixtures. Co-grinding method could improve the dissolution of ROF-β-CyD. However; at 90 minute only 55.98% of the drug was released. This behavior suggests that the dissolution rate increase may be due to reduction in crystallinity of the powder and wetting effect of β-CyD these results are in full accordance with DSC and X-ray data.

Fig. (9) shows the dissolution profiles of ROF powder, physical mixture coevaporate and ground mixture with HP-β-CyD in a 1:1 molar ratio at pH 7.4. The dissolution rate of ROF when physically mixed with HP-β-CyD is two fold higher than that of the untreated drug. This may be due to a local solubilization action operating in the micro-environment or the hydrodynamic layer surrounding the drug particles in the early stages of the dissolution process. This type of cyclodextrin dissolves in a short time thus improving the wettability of the drug, and hence the dissolution of the drug particles[23&27] In other hand the coevaporate and co-ground mixtures of ROF-HP-β-CyD in a 1:1 molar ratio gave 44.6% and 73.1% drug released after 90 minutes respectively. This is may be due to the reduction in crystallinity and partial complexation process. This is in accordance to DSC and X-ray results. Moreover, the extent of the dissolution rate-enhancing effect was found to be dependent on the method used for the preparation of the mixture. So, the most effective preparation method is co-grinding method which could improve the dissolution rate of the drug. It has been reported that grinding of certain drugs with CyD yielded an inclusion complex which exhibited better drug dissolution.[23&28]

Tables (2, 3) show the relative dissolution rate and $t_{50\%}$ for ROF with either β-CyD or HP-β-CyD, respectively; it was found that ground mixture of ROF with HP-β-CyD has a $t_{50\%}$ = 7 min in comparison to ground mixtures with β-CyD ($t_{50\%}$ = 50 min). In other words, ROF-HP-β-CyD

displays a dissolution rate higher than that ROF-β-CyD from their ground mixtures. This implies the solubility enhancing effect of cyclodextrins hence, HP-β-CyD increases the aqueous solubility of the drug than β-CyD as demonstrated in solubility study. Also, the hydroxylation of β-CyD leading to an increase in the hydrophilicity of the parent β-CyD and hence increasing its wettability and aqueous solubility. McCnadless and Yalkowsky.,[29] found that the effect of HP-β-CyD on the dissolution rate of miconazole was higher than that of β-CyD and attributed to the higher water solubility of HP-β-CyD over β-CyD

Table 2: $t_{50\%}$* and RDR** values for ROF / β-CyD systems

System / **RDR at (min)	Plain drug	Physical mixture	Coevaporate	Ground mixture
30	1.00	2.00	6.36	8.04
60	1.00	1.77	5.01	6.45
90	1.00	2.00	5.68	6.70
*$t_{50\%}$	> 90	> 90	>90	50

Table 3: $t_{50\%}$* and RDR** values for ROF / HP-β-CyD systems

System / **RDR at (min)	Plain drug	Physical mixture	Coevaporate	Ground Mixture
30	1.00	2.50	6.35	11.41
60	1.00	2.10	5.54	8.47
90	1.00	2.33	6.00	8.84
*$t_{50\%}$	> 90	> 90	>90	7

*$t_{50\%}$: is the time at which 50% of ROF has been dissolved (i.e. dissolution half – life).

**RDR: Relative dissolution rate: which is the ratio of the amount of the drug released from the prepared systems divided by the amount dissolved from the drug alone at the same time interval.

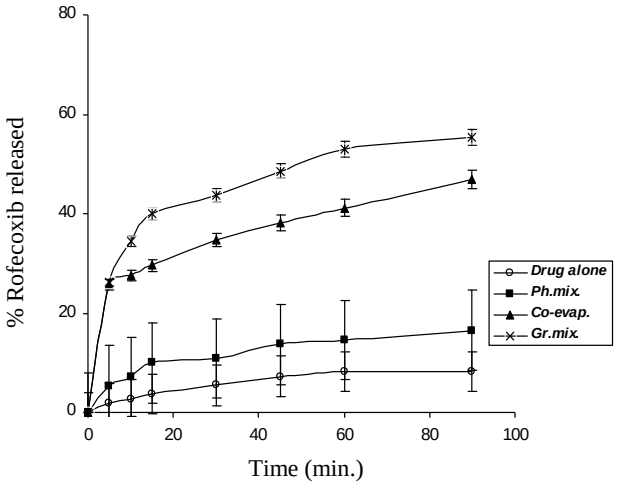


Fig. 8: Release profiles of ROF from different systems containing β-cyclodextrin in phosphate buffer at pH 7.4

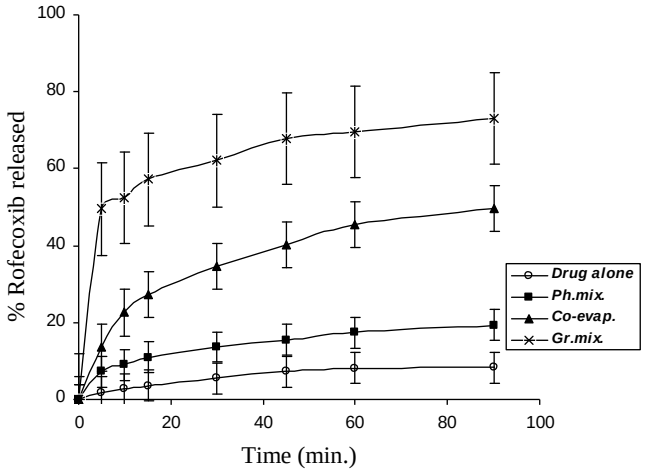


Fig. 9: Release profiles of ROF from different systems containing HP-β-cyclodextrin in phosphate buffer at pH 7.4.

Conclusion

The obtained results indicate that: The solubility of ROF was improved by forming inclusion complexes with β-CyD and HP-β-CyD. Co-grinding technique form partial inclusion complex between ROF with HP-β-cyclodextrin as revealed by DSC, X-ray diffraction and IR studies. β-CyD and HP-β-CyD are useful in enhancing the dissolution rate of water insoluble ROF by using coevaporate and co-grinding techniques. The co-grinding technique is suitable for industrial scale production, allowed us to use this method as the most adequate for the preparation of the inclusion complex of ROF with HP-β-CyD.

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