

STUDIES ON CILOSTAZOL AND β -CYCLODEXTRIN INCLUSION COMPLEXES

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ABSTRACT: The present study deals with studies on Cilostazol & β -cyclodextrin(β CD) inclusion complexes. The present research work describes formation of Cilostazol & β -cyclodextrin complexes using Kneading, Coevaporation & physical mixture with molar proportion of β -CD (1:1). Cilostazol, 6-[4-(1-cyclohexyl-1H-tetrazol-5-yl)-3,4-dihydro-2(1H)-quinolinone] is a vasorelaxant drug & act as a platelet aggregation inhibitor. Cilostazol exhibits low aqueous solubility, which accounts variability in oral bioavailability. Hence, the main objective of study was to investigate the possibility of improving the solubility & dissolution rate of Cilostazol via complexation with β -CD. Phase solubility studies revealed the existence of 1:1 complex between cilostazol and β -CD. Prepared inclusion complexes were characterized by I.R, UV. In vitro release of Cilostazol from inclusion complexes were studied by dissolution study in 0.1N HCl, pH 6.8 and distilled water. It was observed that complexes exhibit higher dissolution rates than the pure drug.

KEYWORDS : cilostazol, β -CD, complexation

INTRODUCTION

Cyclodextrins are oligosaccharides which have received increasing attention in the pharmaceutical field because of their ability to form inclusion complexes with many lipophilic drugs, thus changing their physicochemical and biopharmaceutical properties¹⁻⁵. Among cyclodextrin, β -cyclodextrin is the most widely studied compound for drug complexation. Cilostazol is a vasorelaxant drug act as a platelet aggregation inhibitor it exhibits low aqueous solubility. Cilostazol is practically insoluble in water. The very poor aqueous solubility of the drug gives rise to difficulties in pharmaceutical formulation and may lead to a variable bioavailability.⁶ The objective of the present study was to investigate the possibility of improving the solubility and dissolution rate of cilostazol via complexation with β -CD. In addition, the physicochemical properties of cilostazol- β -CD systems were also investigated and results are reported here.

EXPERIMENTAL

MATERIALS

Cilostazol was a generous gift from Glenmark Pharmaceutical, Ankleshwar. β -cyclodextrin were purchased from Gangwal chemicals, Mumbai. All

reagents were of analytical reagents grade. Double distilled water was used for all the experiments

METHODS

Phase Solubility Studies

Phase solubility studies were carried out at room temperature, in triplicate according to method reported by Higuchi and Connors^{7,8}. Excess amount of cilostazol was added to double distilled water containing various concentration of β -CD (0.002-0.1M) in a series of stoppered conical flasks and shaken for 24 hr on a rotary flask shaker. The suspensions were filtered through Whatman filter paper and assayed for cilostazol using UV spectrophotometer (Varian Cary 100, Australia) at 257 nm against blank prepared using same concentration of β -CD in double distilled water. The association constant (K_a) was calculated from the slope of the linear portion of the phase solubility diagram. According to equation (1),

$$K_a = \text{Slope} / S_0(1 - \text{Slope}) \dots \dots \dots (1)$$

Where, S_0 is aqueous solubility of cilostazol.

SPECTROSCOPIC STUDIES^{9,10}

Complex formation between cilostazol and β -CD was studied by UV spectroscopic method. 10mg cilostazol were weighed accurately and dissolved in 100ml methanol. Diluted suitably and spectra of drug recorded at 257 nm. Same method was used only cilostazol- β -CD complex equivalent to 10 mg of cilostazol were weighed accurately and dissolved in 100ml methanol. Diluted suitably and spectra of complexes were recorded at 257 nm. The change in the absorbance of drug in the complexes was recorded.

PREPARATION OF SOLID COMPLEXES¹¹⁻¹⁵

The solid complexes of Cilostazol & β -CD (1:1 molar ratio) were prepared by following method-

A] Kneading method: (Kn)

Cilostazol & β -CD triturated in a mortar with a small volume of water-methanol solution. The thick slurry was kneaded for 45 min & then dried at 40 °C. Dried mass was pulverized & sieved through (#100)

B] Coevaporation method

The aq. solution of β -CD was added to an alcoholic solution of Cilostazol. The resulting mixture was stirred for 1 hr & evaporated at a temp of 45 °C until dry. The dried mass was pulverized & sieved through (#100)

C] Co-grounding

Drug was triturated with minimum quantity of methanol in a glass mortar until it dissolved. Then β -CD was added and suspension was triturated rapidly at room temperature until solvent evaporated.

D] Freeze -Drying Method

Physical mixtures of Cilostazol & β -CD in a molar ratio of 1:1 was added to 500 ml double distilled water and stirred for 5 days. The suspension was freeze-dried (Ilsin® freeze Dryer), and obtained freeze-dried complex was pulverized and sieved through (<38 μ m)

E] Physical mixture: (PM)

The physical mixtures of Cilostazol & β -CD [1:1 molar ratio] obtained by mixing pulverized powder (#100) together in pestle & mortar.

CHARACTERIZATION OF SOLID COMPLEXES

Saturation Solubility Studies¹⁶

The saturation solubility study was carried out to determine increase in the solubility of pure cilostazol as compared with the physical mixture (PM) and inclusion complexes. Weighed amount of drug, PM and inclusion complexes were added to the 250 mL conical flasks containing 25 mL of double distilled water. The sealed flasks were shaken for 24hr at room temperature followed by equilibrium for three days. Then, the aliquots were withdrawn through Whatman filter paper. The concentration of Cilostazol was determined by UV spectrophotometer at 257 nm.

Percentage yield

Percentage yield is calculated to know about per cent yield or efficiency of any method,

$$PY (\%) = \frac{\text{Practical Mass (Solid dispersion)}}{\text{Theoretical Mass (Drug + Carrier)}} \times 100$$

Drug content

Cilostazol- β -CD complex equivalent to 10mg of Cilostazol were weighed accurately and dissolved in 100ml of methanol. Diluted suitably and drug content was analyzed at 257nm by UV spectrophotometer UV spectrophotometer (Varian Cary 100, Australia) at 257 nm.

FT-IR Spectroscopy

IR spectra of pure Cilostazol, β -CD and with its complexes were obtained by a Varian 640 IR spectrophotometer (Varian, Australia), using KBr pellets. The scanning range used was 4000 to 400 cm^{-1} .

In-vitro dissolution studies

Dissolution studies of samples were performed according to USP XXIII type II apparatus 900mL of 0.01N HCl, pH 6.8, double distilled water. The temperature was maintained at 37 \pm 0.5 °C and the rotation speed was 75 rpm. The samples were withdrawn at time intervals 5, 10, 15, 20, 30, 45, 60 and 90 minutes and analyzed spectrophotometrically at 257nm.

RESULTS AND DISCUSSIONS

The phase solubility diagram for the complex formation between cilostazol and β -CD is shown in Fig.1. The aqueous solubility of cilostazol increased linearly with a slope 0.9269 ($r^2 = 0.9921$), as a function of β -CD concentration. The phase solubility diagram can be classified as type A according to Higuchi and Connors. It is assumed that the increase in solubility observed was due to the formation of a 1:1 M inclusion complex. The association constant (K_a) was calculated as 468.93 M^{-1} which was within range of 200-5000 M^{-1} . The UV spectra of cilostazol solution in the presence of β -CD were studied. An example is shown in Fig.2. There was no shift in the λ_{max} of cilostazol in the presence of β -CD. The spectra of complexes showed a diminution in the absorbance at 257 nm. The induced change in absorbance is attributed, primarily, to the formation of an inclusion complex. The change in peak intensity are assumed to result from changes in the solvent microenvironment upon inclusion of the solute. The observed reduction in peak intensity may result from the transfer of the guest molecule from water to the CD cavity. The saturation solubility data for PMs and complexes of cilostazol- β -CD are presented in Table 1. The solubility showed a steep increase from 10.2 $\mu\text{g/ml}$ to 21.88 $\mu\text{g/ml}$.

The results of percent practical yield studies are shown in Fig.3 the maximum yield was found to be 97.50% in complex prepared by Freeze-drying method. Percentage drug content of the complexes are shown in Fig. 4 was found within the range of 93.90 \pm 0.5 to 96.71 \pm 0.5

Fig.3 illustrates the FTIR spectra of the samples under study. IR spectrum of Cilostazole shows prominent peaks at 3329, 2922, 1668, 1155, 750 cm^{-1} corresponding to N-N stretching in amine, C-H stretching in alkane, C=O stretching in aryl ketone, ether and C-H bending in aromatic ring. FT-IR spectra of Cilostazole, β -CD, physical mixture, and Cilostazole- β -CD complexes were obtained indicating no chemical interaction between the

drug and β -CD which confirmed the stability of the drug with its complex. Fig. 6, Fig7, Fig8 From in vitro release study, it was found that the complex prepared as 1:1 by Freeze-drying method has shown improve in dissolution behavior as compaire to drug and other complexes. Which indicate the improved solubility phenomenon.

CONCLUSIONS:
Cilostazol- β -CD complex(1:1) prepared by Freeze-drying method showed increases in solubility and dissolution rate in comparison with plain drug and other complexes. This technique would be used to develop fast release formulations of Cilostazole.

Table1 :Saturation Solubility Data of Inclusion Complexes in Double Distilled Water

SDs	Solubility (mcg /ml)	% increase solubility
Drug	10.02
A	20.51	204.72%
B	20.93	208.88%
C	19.1	190.65%
D	21.88	218.36%
E	18.22	181.83%

Where Drug- Cilostazol, A-Kneading method:(Kn), B-Coevaporation method, C- Co-grounding , D- Freeze –Drying Method, E- Physical mixture: (PM)

Fig.1 Phase solubility diagram of cilostazol β -CD

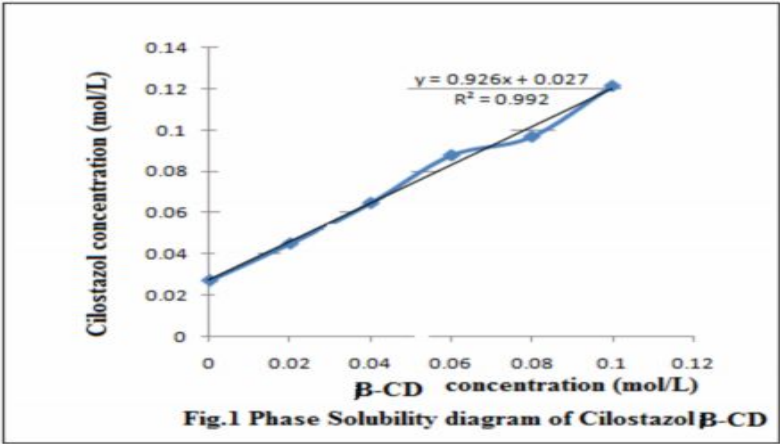


Fig.2 Effect of β -CD concentration on UV absorption of cilostazol

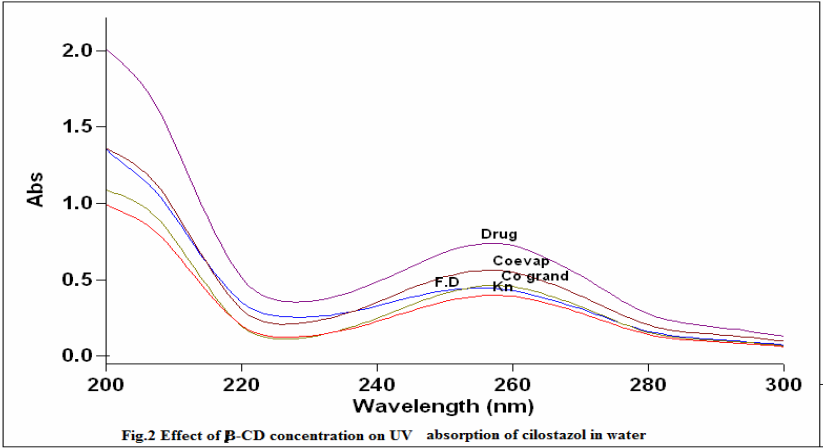


Fig.3 % practical yeild

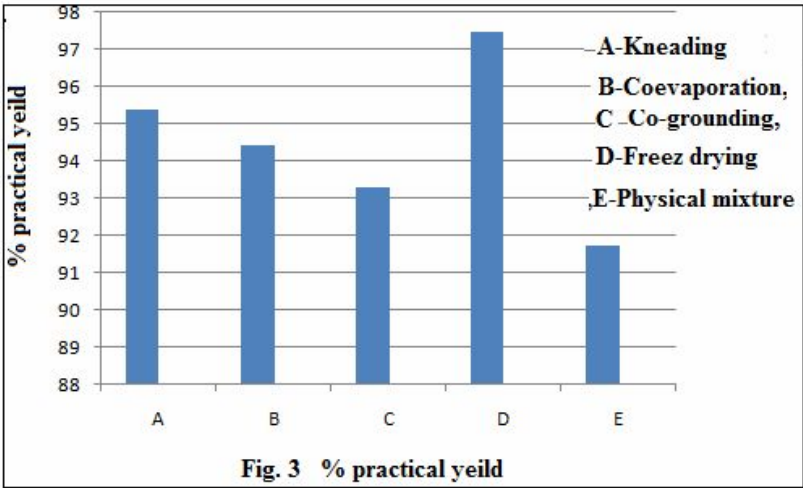


Fig.4. % drug content

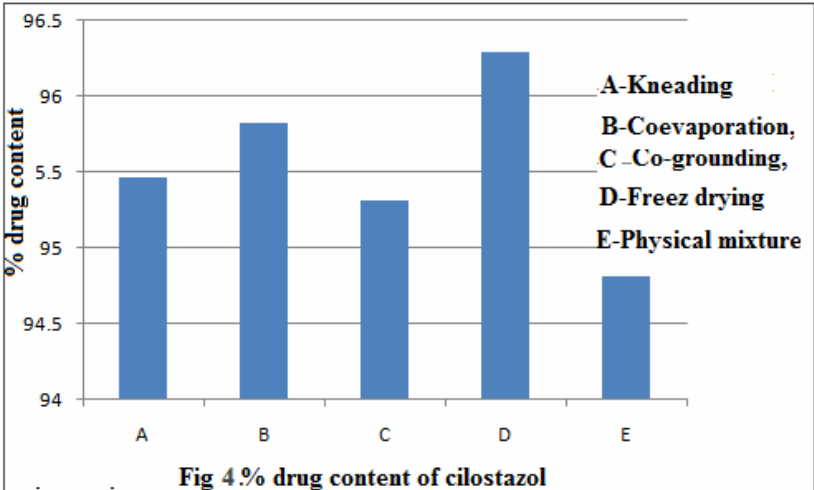


Fig.5. FTIR Spectra of cilostazole and complex

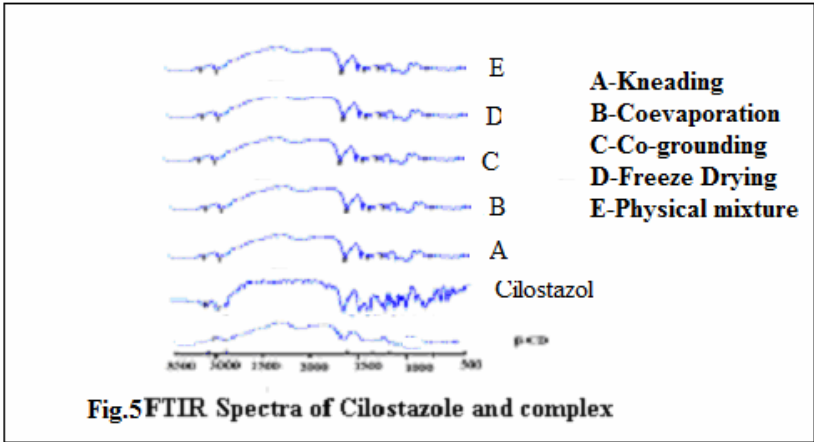


Fig.6.In-vitro dissolution profile in 0.1N HCl

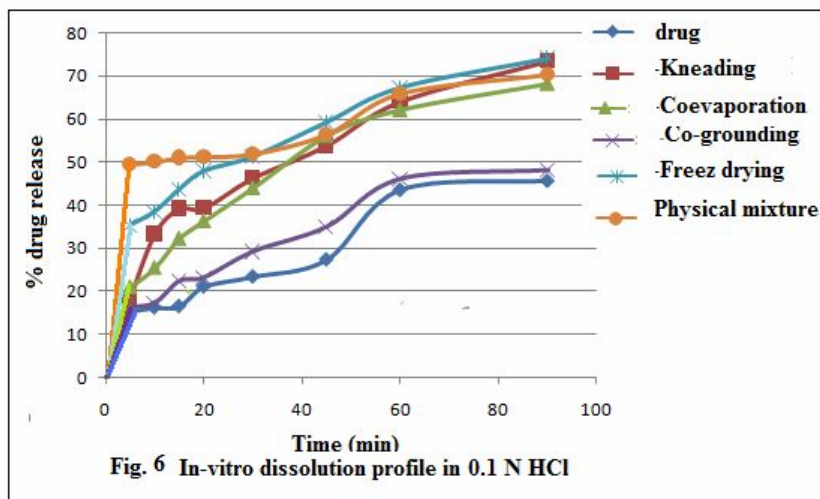


Fig.7. In-vitro dissolution profile in pH6.8

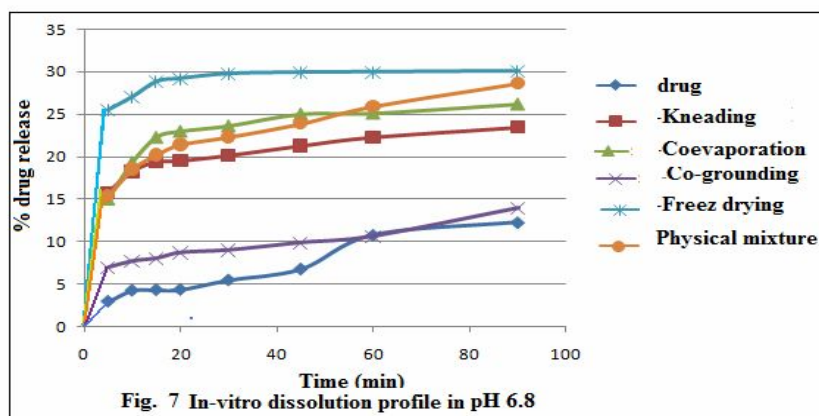
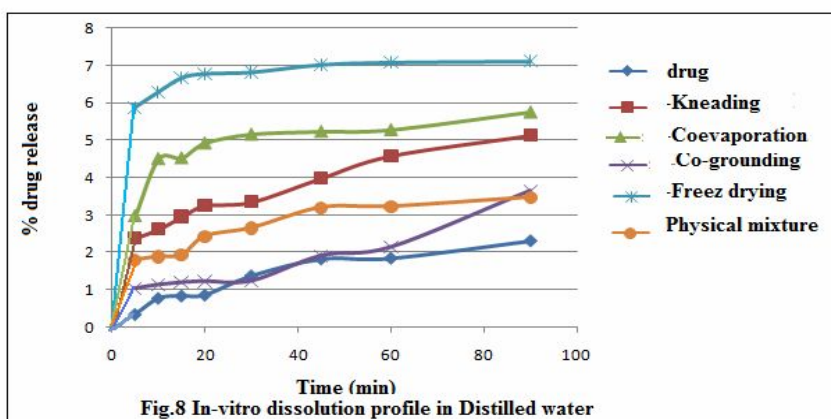


Fig.8.In-vitro dissolution profile in Distilled water



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