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Preparation and characterization of rufinamide HP- β -cyclodextrin complexes prepared by the kneading method for solubility enhancement.

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Original Paper Artículo Original

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RESUMEN

Objetivos: La presente investigación se refiere a la preparación y caracterización de complejos de rufinamida HP- β -ciclodextrina preparados por el método de amasado.

Material y métodos: La rufinamida fue donada por la empresa Torrent Pharmaceuticals limitado. HP- β -ciclodextrina (HP- β -CD) se adquirió de Himedia, India. Metanol y ácido clorhídrico se obtuvieron de SD Fine Chem. SA. Ltd., India. Se utilizó el método de amasado para preparar complejos de inclusión de rufinamida. El estudio de la fase de solubilidad se realizó para comprobar la formación de complejos de inclusión. Los complejos preparados se caracterizaron por diferentes métodos como DSC, FTIR, X-RPD y ensayo de disolución in vitro.

Resultados: Se encontró que se producían formación de complejos en la relación 1:1. La constante de estabilidad encontrada fue de 221,27 M⁻¹. Los estudios de DSC, FTIR confirmaron la formación del complejo de inclusión. Mediante X-RPD se confirmó la naturaleza amorfa del complejo.

Conclusiones: El estudio de disolución in vitro mostró que la proporción 1:1.5 liberaba alrededor de 50% de fármaco en 30 min y a los 60 minutos se consiguió una liberación del 70%.

PALABRAS CLAVE: Hidroxi propil β -ciclodextrina, método de amasado, rufinamida, DSC, FTIR, X-RPD.

ABSTRACT

Aims: The present investigation concerns the preparation and characterization of Rufinamide HP- β -cyclodextrin complexes prepared by the kneading method.

Material & methods: Rufinamide was procured as a gift sample from Torrent Pharmaceuticals limited. HP- β -cyclodextrin (HP- β -CD) was purchased from Himedia, India. Methanol and Hydrochloric Acid were purchased from S. D. Fine Chem. Pvt. Ltd., India. kneading method was selected to prepare inclusion complexation of Rufinamide. Phase solubility study was performed to check formation of inclusion complex. Prepared complex were characterize by different methods like DSC study, FTIR study, X-RPD study & in-vitro dissolution study

Results: It was found that there is a formation of 1:1 inclusion complex between HP- β -CD as stability constant was found to be 221.27 M⁻¹. DSC study, FTIR study had given supporting data for formation of inclusion complex. Amorphous nature of the complex was confirmed from the X-RPD study.

Conclusions: From in-vitro dissolution study it was found that 1:1.5 complex showed around 50% drug released in 30 min & more than 70% of Drug release in 60 mins.

KEY WORDS: Cyclodextrins, Differential Scanning Colorimetry, Hydroxy propyl β -cyclodextrin, kneading Method, Rufinamide

INTRODUCTION

Rufinamide (RUF) [1-(2, 6-difluorophenyl) methyl-1H-1, 2, 3-triazole-4-carboxamide] is a triazole compound and has a chemical formula of $C_{10}H_8F_2N_4O$. In experimental models, RUF has been shown to modulate sodium channels, prolonging their inactivation phase, thereby limiting the firing of sodium-dependent action potentials in neurons, and resulting in a membrane stabilizing effect.^{1,2} However, it's very low aqueous solubility around 40 mg/L³ and poor dissolution can cause formulation problems and limit its therapeutic application by delaying the rate of absorption and the onset of action.⁽²⁾ Solid dispersions of Rufinamide were reported using HPMC by solvent evaporation method for improving dissolution.⁽⁴⁾ Cyclodextrins are commonly used in drug formulations as solubility enhancers because of their ability to form water soluble inclusion complexes with poorly water soluble drugs.⁽⁵⁾ The method of complexation may play a role in drug solubilization.⁽⁶⁾ Therefore, it seemed of interest to extend our investigations to a series of binary systems of Rufinamide with HP- β -CD.

METHODS

Materials

Rufinamide drug was procured as a gift sample from Torrent Pharmaceutical Ltd., India. HP- β -cyclodextrin (HP- β -CD) was purchased from Himedia, India. Methanol and Hydrochloric Acid were purchased from S. D. Fine Chem. Pvt. Ltd., India.

Calibration Curve

1 mg/ml stock solution of drug was prepared in methanol. All further dilutions to make 50, 75, 100, 125, 150 μ g/ml standard solutions were made by 0.1N HCl. The spectra of the standard solutions were recorded using UV Visible spectrophotometer for 200nm to 400nm range against 0.1N HCl as blank. The observations were recorded in triplicate.

Phase solubility studies

An excess of drug was added to 5 mL of HP- β -CD aqueous solutions (5–25 Mmol) in 10-mL stoppered conical flasks and shaken at room temperature in an orbital shaker incubator.⁽⁷⁾ At equilibrium after 48 hours, aliquots were withdrawn, filtered (0.22 μ m pore size, Whatman) and assayed for drug content at 262.0 nm using uv visible spectrophotometer (UV-1800, Shimadzu, Japan). The apparent 1:1 stability constant of the Rufinamide-cyclodextrin complex was calculated from the phase-solubility diagram:

$$Kc = \frac{\text{Slope}}{S_0(1 - \text{Slope})}$$

where Kc is the stability constant (L mol⁻¹), slope is obtained from the linear /relationship between the concentration of Rufinamide and HP- β -CD and S₀ is aqueous solubility of Rufinamide (mmol⁻¹ L).

Preparation of physical mixtures and the solid inclusion complex

For physical mixtures, RUF and HP- β -CD were weighed accurately at a 1:1 molar ratio, mixed thoroughly by trituration in a mortar and sieved through 0.25 mm sieve. All physical mixtures were stored in dessicator until further evaluation. The inclusion complex of Rufinamide with HP- β -CD was prepared at a 1:1 molar ratio by wetting the physical mixture in a mortar with a minimum volume of ethanol/water (1:1, by volume) mixture and kneading thoroughly for 60 min with a pestle to obtain a paste, which was then dried under tray drier at 50°C, sieved through 0.25 mm sieve and stored in a dessicator until further evaluation.

Dissolution studies^(8,9)

Dissolution studies were performed in 0.1 N HCl (pH 1.2, 900 mL) at 37 \pm 0.2 °C, using USP XXIII apparatus (Electrolab, India) with a paddle rotating at 50 rpm. Solid products, each containing 100 mg of drug, were subjected to dissolution. At fixed time intervals (5, 10, 15, 30, 45 and 60 minutes), withdrawn samples were filtered (Whatman filter paper No. 41) and assayed for drug content at 262.0 nm using uv visible spectrophotometer (UV-1800, Shimadzu, Japan).

Differential scanning calorimetry (DSC)⁽¹⁰⁾

DSC thermograms of the drug, cyclodextrin, physical mixture and kneaded complex were recorded on a Perkin Elmer 1/ DSC equipment, Massachusetts, USA. The instrument was calibrated with indium and zinc prior to analyzing the samples under nitrogen. All accurately weighed samples (2.5 mg) were placed into sealed aluminium pans and scanned at the heating rate of 10 °Cmin⁻¹ over the temperature range of 30–240 °C.

X-ray Diffractometry⁽¹¹⁾

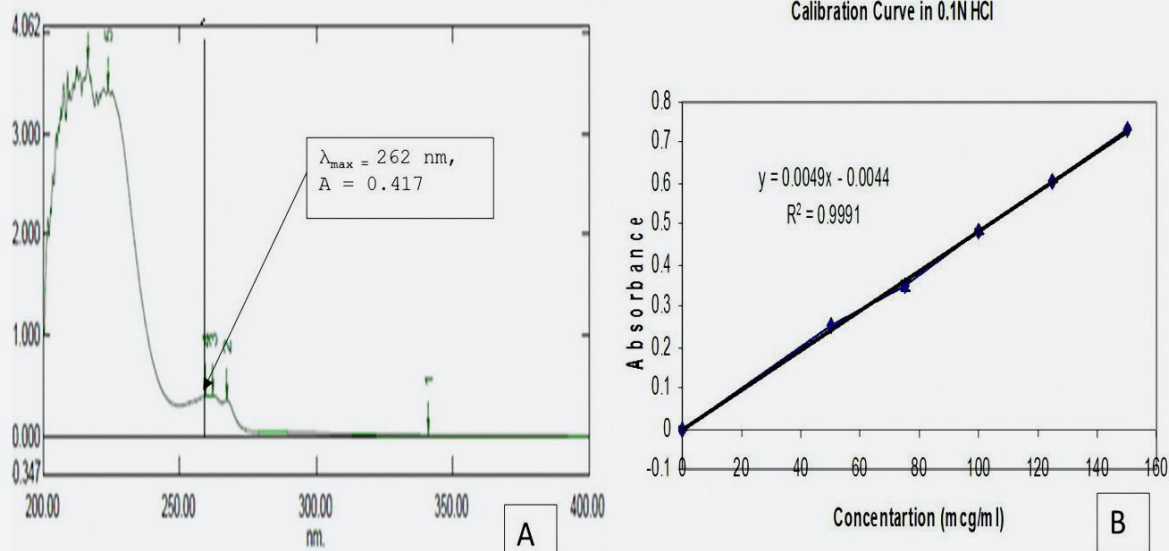
X-ray powder diffraction patterns were recorded using D2 Phaser, BRUKER, AXS Inc. Germany at 40mV, 45 kV and with monochromatized Cu K α radiation (λ = 1.54056Å). The samples were scanned at room temperature in the continuous scan mode over the 10⁰–80⁰ range.

RESULTS AND DISCUSSION

Preliminary studies

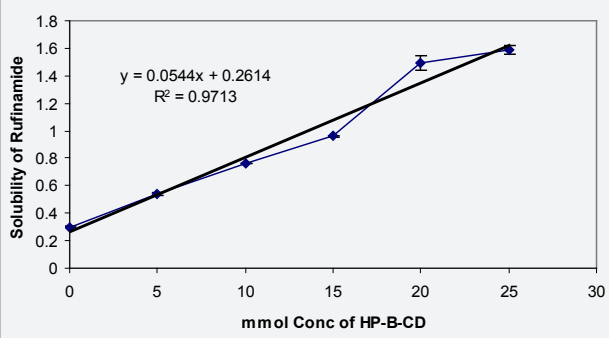
The Complexation of Rufinamide with HP- β -CD was studied at molar ratios of 1:1, 1:1.5, 1:2. The complexes were

Figure 1: UV spectra & Calibration curve of drug in 0.1N HCl.



A: UV spectra of drug; B: Calibration curve of drug in 0.1N HCl

Figure 2. Phase solubility Diagram



prepared by the kneading method and were characterized by DSC, FTIR and X-RAY Diffraction studies.

All complexes showed increased dissolution efficiency, reduced time required for 50% drug to be dissolved higher dissolution (> 50% released in 30 min) was obtained using a 1:1.5 molar ratio of the drug to HP-β-CD. Therefore further investigations were carried out for the 1:1.5 molar ratios.

Calibration Curve

The drug spectra (Figure 1-A) showed a prominent peak at 262 nm which indicates that trace amount of methanol did not interfere in absorption. It was concluded that the ranges of absorbance lies in ranges of 0.2 to 0.8 and hence it follows Lambert's beers law. The R^2 value was found to be 0.9991 (Figure 1-B).

Phase solubility

Rufinamide is a practically insoluble drug. The solubility of rufinamide in distilled water at room temperature was 32.0 mg/L and was notably affected by the presence of HP-β-CD. The solubility of rufinamide is linearly increases with increase in the concentration of HP-β-CD from 0-25 mmol concentration. The obtained phase solubility diagram was linear (Figure 2) and could be classified as A_L type according to Higuchi and Connors. The slope of solubility diagram was less than one; it was therefore assumed that the solubility increase could be attributed to the formation of the 1:1 complex. Stability constant value obtained for the

Table 1: Comparison of results of dissolution of pure drug with inclusion complex

Time (min)	% DRUG RELEASE [AVERAGE ± S.D. (N=3)]			
	Pure Drug	Complex (1:1)	Complex (1:1.5)	Complex (1:2)
5	12.20±0.66	24.69±3.67	27.13±4.62	28.73±5.55
10	14.84±1.08	31.08±4.66	34.78±5.66	31.13±5.67
15	15.62±0.86	38.16±7.44	40.68±6.56	48.00±7.55
30	16.28±0.49	45.92±9.36	50.92±4.80	53.42±5.71
45	18.29±0.68	54.99±6.72	60.65±5.84	57.68±3.94
60	18.79±0.50	66.00±5.95	74.17±4.92	69.93±4.92

Figure 3: Comparison of dissolution profile of pure drug with complexes

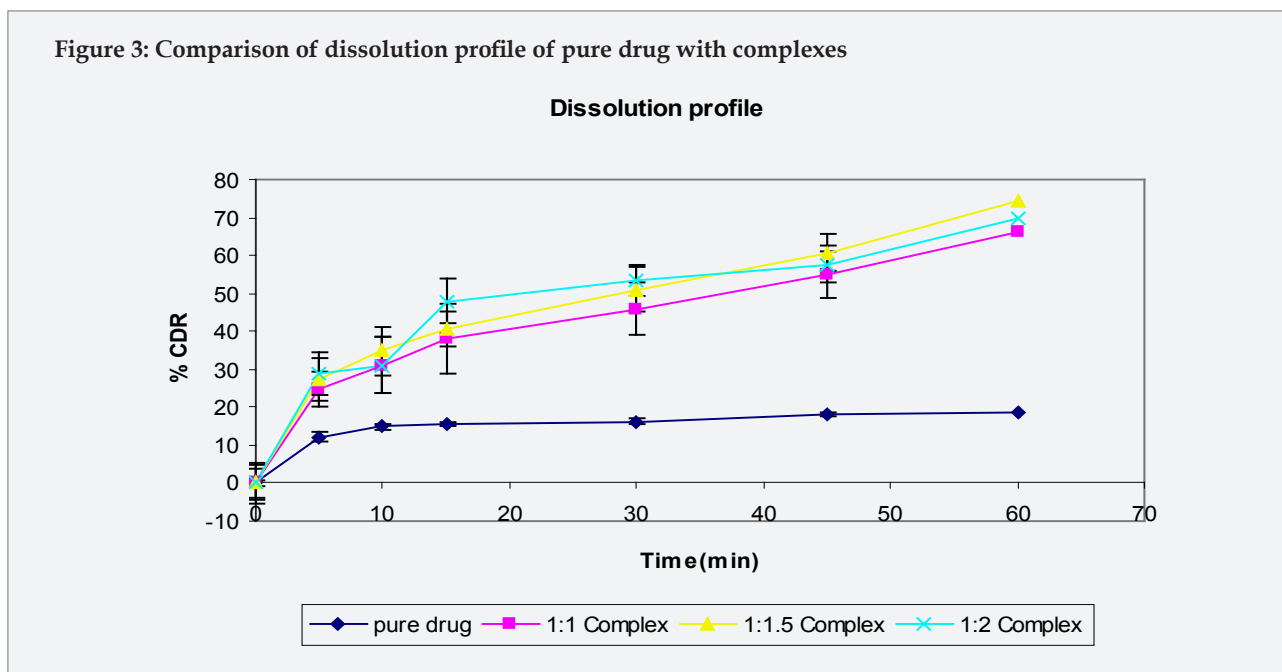
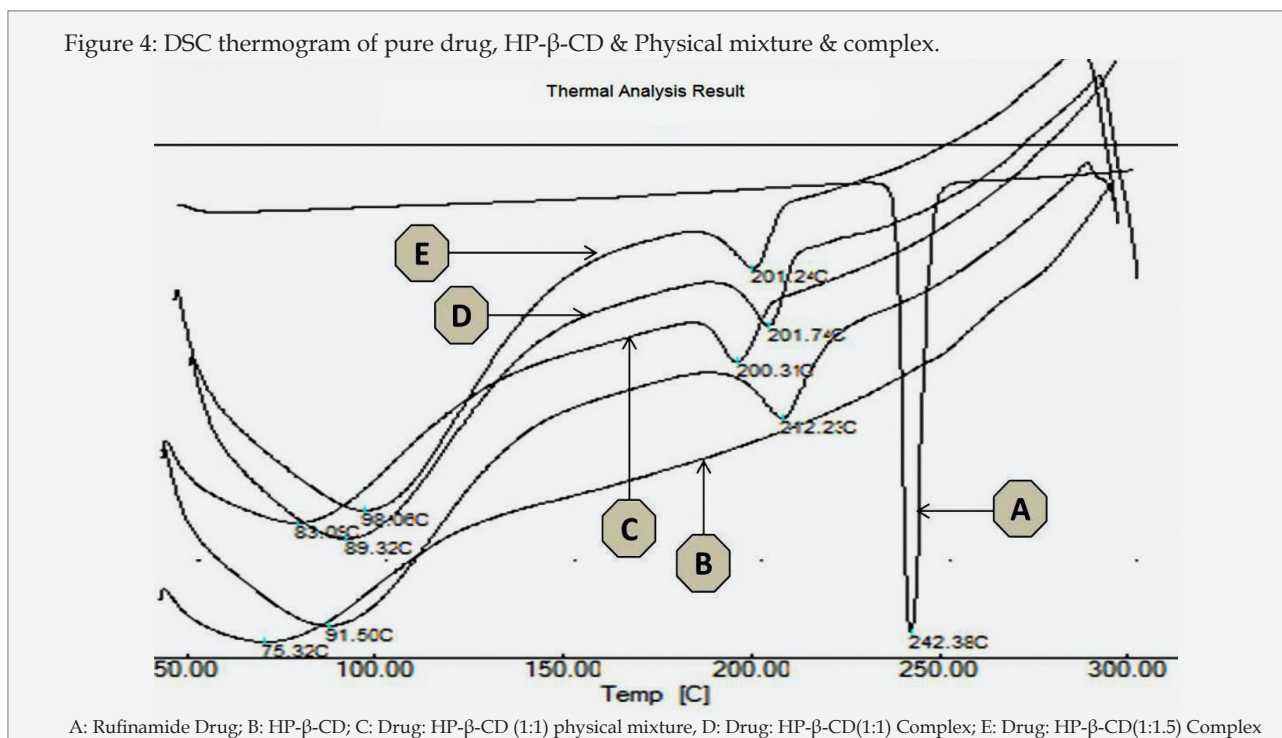


Figure 4: DSC thermogram of pure drug, HP-β-CD & Physical mixture & complex.



Rufinamide-HP-β-CD complex was 221.27 M^{-1} which well within the ideal range of 100 M^{-1} to 1000 M^{-1} . The K_c value indicated that the Rufinamide-HP-β-cyclodextrin complex at a 1:1 ratio is adequately stable.

Dissolution study

The in vitro dissolution profiles of the pure drug and inclusion complex of 1:1, 1:1.5 & 1:2 ratios were shown in figure 3. The dissolution of complexes was higher compared to the drug alone. The dissolution profile of the kneaded complex 1:1.5 molar ratio showed 74.17% drug released in

60 min while the pure drug showed 18.79% (Table 1). This enhancement can be attributed to the higher hydrophilic character of the systems due to the presence of the carrier, which can reduce the interfacial tension between the poorly water soluble drug and the dissolution medium.

Moreover, in the case of HP-β-CD, in the early stage of the dissolution process, the carrier dissolves more rapidly than the drug. Hence, it can act on the hydrodynamic layer surrounding the drug particles, resulting in an in situ inclusion process that improves the dissolution of the drug.

In fact, the systems containing a larger amount of HP- β -CD showed faster drug dissolution. As shown above, the kneaded complex showed 4 time higher dissolution in 60 min than that of pure drug.

DSC study

DSC curves of the pure drug, HP- β -CD, physical mixture, 1:1, 1:1.5, 1:2 molar kneaded complex are shown in figure 4. DSC curve of the drug showed a sharp endothermic peak at 242.38°C the sharp melting peak of rufinamide was shifted towards 200.31°C, 201.74°C in case of 1:1 & 1:1.5 respectively. Shifting of HP- β -CD peak towards 89.32°C, 83.09°C of 1:1 & 1:1.5 respectively, this was a deviation from 75.32°C of pure HP- β -CD. So this change indicates formation of inclusion complex.

X-RAY Diffraction Study

From the X-RD data it was cleared that the drug was in crystalline state it was confirmed from the angle of diffraction because peak position is an indication of crystal structure in which peak height is a measure of crystallinity. Diffractogram of pure drug exhibited characteristic intense peaks at 2θ value of 10° to 30° . They were reduced after inclusion Complexation formation with HP- β -CD prepared by kneading method. So it is indicating of amorphous state of a pure drug. This was the confirmation of the change in state of rufinamide. So this might be a reason for increase in the solubility of rufinamide after inclusion Complexation.

CONCLUSIONS

The dissolution profile of the kneaded complex (1:1.5 molar ratio) showed more than 70% drug released in 60 min. Further study is required to prepare a prompt release oral formulation of rufinamide by utilizing this complex.

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