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COCRYSTALS OF RAMIPRIL FOR ENHANCEMENT OF SOLUBILITY AND DISSOLUTION PROPERTIES

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ABSTRACT
This research work was intended to improve the solubility and dissolution properties of Ramipril by the development of bicomponent cocrystals. Bicomponent cocrystals of Ramipril were prepared with β-cyclodextrine cocrystal former (CCF) by neat grinding techniques. Solubility analysis of cocrystals showed that Ramipril cocrystals have comparatively better solubility than the other cocrystals. The solubility of pure Ramipril (257.23 µg/mL) was found to be improved by seven folds (1799.61 µg/mL) due to the formation of bicomponent crystalline form with β-cyclodextrine. *In vitro* dissolution also revealed that the immediate release tablet of Ramipril showed a release of 59.60±1.31%, while Ramipril cocrystal tablet showed drug release of 95.65±1.21 % after 60 minutes. Results of Differential Scanning Colorimetry (DSC), Powdered X-Ray Diffraction (PXRD) and Raman spectroscopic analysis also justified the formation of the novel crystalline form. The developed bicomponent cocrystals of Ramipril improved the solubility and dissolution of Ramipril to a significant extent.

KEYWORDS: Solubility analysis of cocrystals showed that Ramipril cocrystals have comparatively better solubility than the other cocrystals.

INTRODUCTION
Ramipril is an angiotensin converting Enzyme Inhibitor, Antihypertensive agent. It is chemically(2S,3aS,6aS)-1-[(2S)-2-[[(2S)-1-ethoxy-1-oxo-4phenylbutan-2-yl]amino] propanoyl] 3,3a, 4, 5,6,6a-hexahydro-2H cyclopenta [d] pyrrole-2-carboxylic acid. Ramiprilat is an active metabolite of Ramipril,competes with angiotensin I for binding at the angiotensin converting enzyme, blocking the conversion of angiotensin I to angiotensin II. As angiotensin II is a vasoconstrictor and a negative feedback mediator for renin activity, lower concerntration results in decrease in blood pressure and an increase in plasma renin. Ramipril is used in the treatment of hypertension, post myocardial infarction, cardiac stroke, and cardiovascular diseases. It is a BCS class –II, pKa 5.2, drug. Elimination half life is 13-17hr. The absorption of the Ramipril after oral administration is 50%-60%. The Ramipril having poor water solubility which results in the low bioavailability i.e. 28% The common side effects arise after administrattion of Ramipril are vomiting, nausea, headache, dizziness, vertigo. To solve the above cited problems, cocrystals of Ramipril is better alternative. Cocrystals of Ramipril have potential advantages over conventional dosage forms, with their improved patient compliance; convenience bioavailability and rapid onset of action had drawn the attention of many manufacturers over a decade. It provides fast dissolution by preparing immediate release tablets.

MATERIALS AND METHODS
Ramipril was received as a gift sample from Ajanta Pharma Aurangabad, and all coformers were procured from Balaji drugs Malegaon..

Preparation of bicomponent crystals (cocrystals)
This bicomponent crystalline system was consisting of drug and cocrystal former (CCF). The neat grinding method was implemented for the preparation of cocrystals. Ramipril and β-cyclodextrin as a coformer (1:1 stoichiometric ratio) were subjected for neat grinding in mortar and pestlefor 30 min and the resulting product was subjected to physicochemical characterization.[11]

Solubility analysis of cocrystals
Higuchi and Connors approach was used to determine the solubility. An excess amount of drug was added to vials containing 10 mL of water. The shaking of vials was carried out using a rotary shaker and was followed by sonication. The resulting solution was allowed to stand for 24 h, and then samples were filtered through Whatman filter paper and the filtrate was subjected to UV Spectrophotometric analysis at the wavelength of 210 nm after proper dilutions.[12] The cocrystals with the highest solubility were characterized using Differential

Scanning Colorimetry (DSC), X-ray Diffraction (XRD), and Raman spectroscopy.[13]

Differential Scanning Colorimetry (DSC) analysis

The cocrystals were thermally analyzed using a Differential Scanning Colorimeter (DSC) (Mettler Toledo). The powder sample (2 mg) in the hermetically sealed aluminium pan was heated at a scanning rate of 20 °C/ min from 50° to 300 °C in constant nitrogen flow of 20 mL/min.[14]

Powder X-ray Diffraction (PXRD) analysis

The PXRD patterns of pure drug, coformer, and cocrystal were recorded at different 2θ values. The analysis was carried out with the 2°/min scanning speed and 2°/2 cm chart speed per 2θ.[15]

Raman spectroscopic analysis

Raman spectroscopic analysis is one of the most important spectroscopic tools in structural analysis. The structural analysis of pure drug and cocrystals was performed using the Fourier Transform Raman spectrometer (Bruker RFS-27) to determine the different principle functional groups.[16]

Flow property study of active pharmaceutical ingredient (API) and cocrystals.

The different micromeritic parameters like bulk density (BD), tapped density (TD), Hausner's ratio (HR), Carr's index (CI), and angle of repose (AR) were determined for pure drug and cocrystals to compare the improvement in flow properties.[17]

Formulation and evaluation of tablets

The immediate-release tablets of plain Ramipril and Ramipril cocrystals were prepared using the direct compression method (Table I). The tablet formulation of Ramipril and Ramipril cocrystal was evaluated for weight uniformity, friability, and hardness. Evaluation of uniformity of weight was done by weighing 20 tablets separately and comparing the average weight of 20 tablets with the weight of the individual tablet. To evaluate the mechanical strength ofthe tablet, the hardness was evaluated using a hardness tester (Veego). The friability of the tablets was determined using a friability tester (Veego) at 25 rpm.[18]

***In vitro* dissolution studies**

USP apparatus type II (Veego) was used for comparative *in vitro* dissolution studies of Ramipril and Ramipril cocrystal tablets. The apparatus was maintained at 37±0.5 ^{0} C and 900 mL water was used as dissolution medium. After an interval of every 10 min a 5 mL sample was withdrawn and repeated up to 60 min. After the withdrawal, every time 5 mL fresh water was added to maintain the sink condition. The samples were analyzed after filtration using a UV 1800 double- beam spectrophotometer (Shimadzu) at the wavelength of 274 nm.[19,20]

Stability studies

To perform a stability study, the tablets of Ramipril cocrystals were packed in aluminium foil and stored at 40 °C ± 2 °C temperature and 75 % ±5 % RH for one month. At the end of the month, the different evaluations like drug content, disintegration time, hardness, and friability were performed.[21]

RESULTS AND DISCUSSION

After the screening of all Ramipril cocrystals prepared with β-cyclodextrin coformer, it was found that cocrystals prepared with β-cyclodextrin were having the maximum solubility. In quantitative terms, the solubility Ramipril in water was 257.23 µg/mL, and the solubility of Ramipril cocrystal was found to be.

Table I: Formulation of Ramipril and Ramipril-βcocrystal tablet.

Sr. No.	Ingredients	Quantity (mg)	
		Ramipril Tablet	Ramipril Cocrystal Tablet
1	Ramipril / Cocrystal	20	27.5
2	Lactose	91.5	84
3	Cross povidone	22	22
4	Avicel pH-101	15	15
5	Magnesium Stearate	1.5	1.5

Table II: Preformulation characteristics of Ramipril and Ramipril-β-cyclodextrin cocrystals.

Properties	Angle of repose (^{0})	Bulk density (g/mL)	Tapped density(g/mL)	Carr's index	Hausner's ratio
Ramipril	41.66 ± 1.45	0.30 ± 0.06	0.52 ± 0.07	32.40 ± 1.17	1.73 ± 0.09
Ramipril Cocrystals	23.77 ± 1.12	0.54 ± 0.09	0.65 ± 0.03	16.92 ± 0.88	1.20 ± 0.05

Mean ± SD, n=3

Table III: Evaluation of Ramipril and Ramipril-β-cyclodextrin tablets.

Batch	Thickness (mm)* (n=3)	Hardness (Kg)* (n=3)	Uniformity of weight (g)* (n=20)	*In vitro* disintegration time (min)* (n=3)	%Friability (%)* (n=20)
Ramipril	5.03 ± 0.03	1.8 ± 0.4	0.35 ± 0.03	15 ± 1.0	0.76 ± 0.30
Ramipril Cocrystals	5.05 ± 0.02	1.7 ± 0.2	0.35 ± 0.02	14 ± 0.5	0.66 ± 0.31

Mean ± SD, n=3

Table IV: Stability study Ramipril and Ramipril-β-cyclodextrin tablets.

Parameters	Thickness (mm)* (n=3)	Hardness (kg)* (n=3)	Drug Content (%)* (n=3)	*In vitro* disintegration time (min)* (n=3)	%Friability(%)* (n=20)
40 ± 2 °C / RH 75±5 %	5.04 ± 0.03	1.6 ± 0.1	99.14 ± 0.77	13 ± 0.3	0.65 ± 0.22

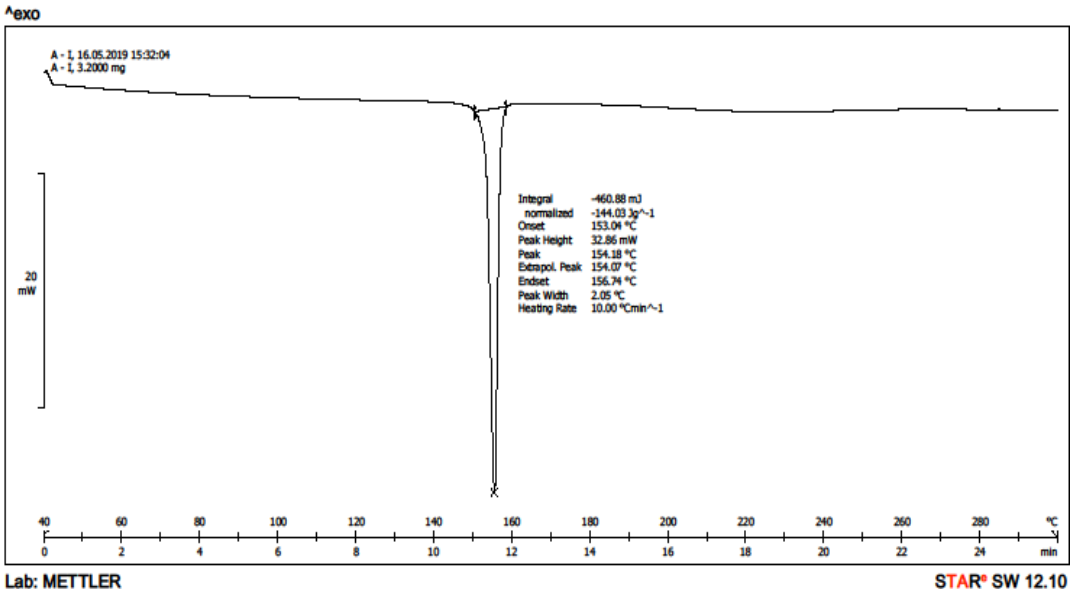
Mean ± SD, n=3

increased by seven folds to that of original and it was1799.61 µg/mL.

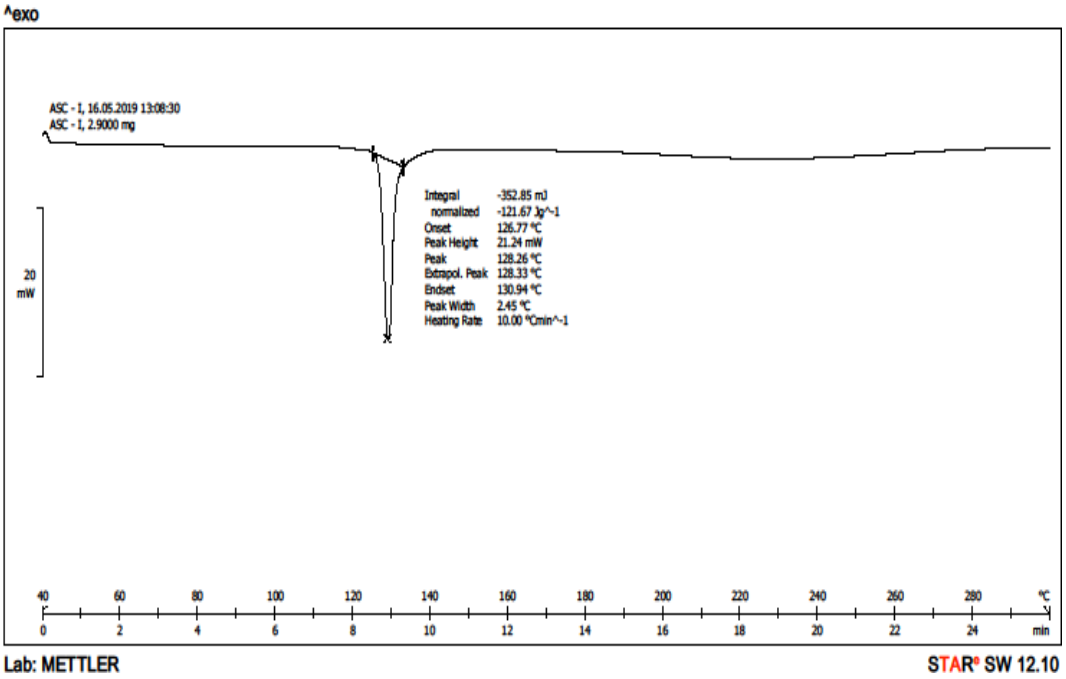
DSC analysis was done to determine the phase of transformation in the formation of cocrystals. DSC thermogram of ramipril (Fig. 1A) showed an exothermic peak at 115.18° corresponding to its melting point. The changes in the peak were observed in the case of the Ramipril cocrystals. The DSC peak for cocrystals was observed at 120.26° (Fig. 1B). Cocrystallization is the result of strong supramolecular interaction between the API and the coformer.[23] The hydrogen bonding between polar functional groups is the major factor in the formation of cocrystals. This interaction between drug and coformer resulted in moderate to complete alterations in crystalline arrangement with significant changes in physical properties as well as melting point and solubility.[24]

The PXRD patterns of pure drug and cocrystals are presented in Fig. 2A and 2B, respectively. The important diffraction peaks for ramipril were detected at different 2θ values. However, in cocrystals, there were changes in diffraction peaks with slight variation in intensity. The appearanceof different and additional diffraction peaks in comparison with drug and coformers reveals the formation of thenew crystalline form.[25]

Raman *spectroscopy is a significant tool for interpreti*ng the formation of.

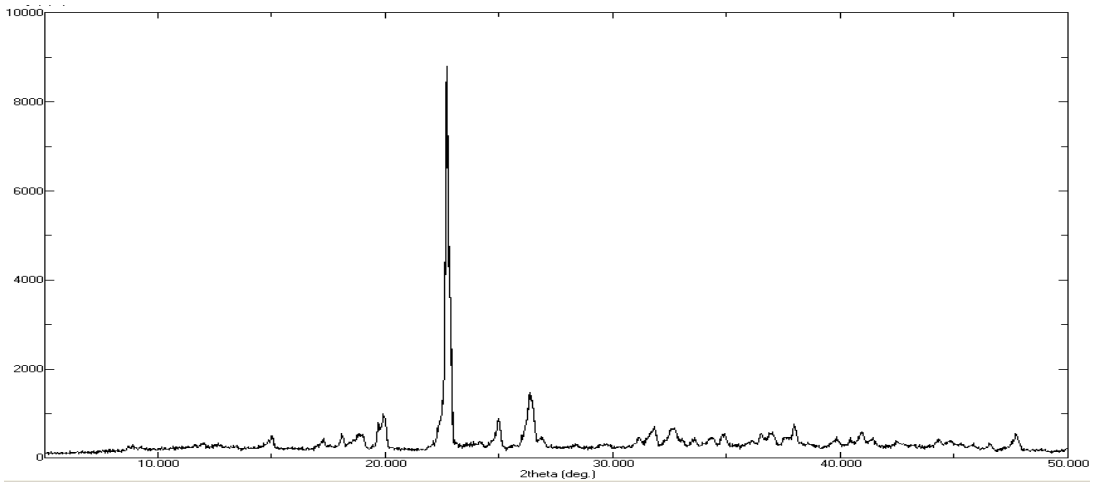


A

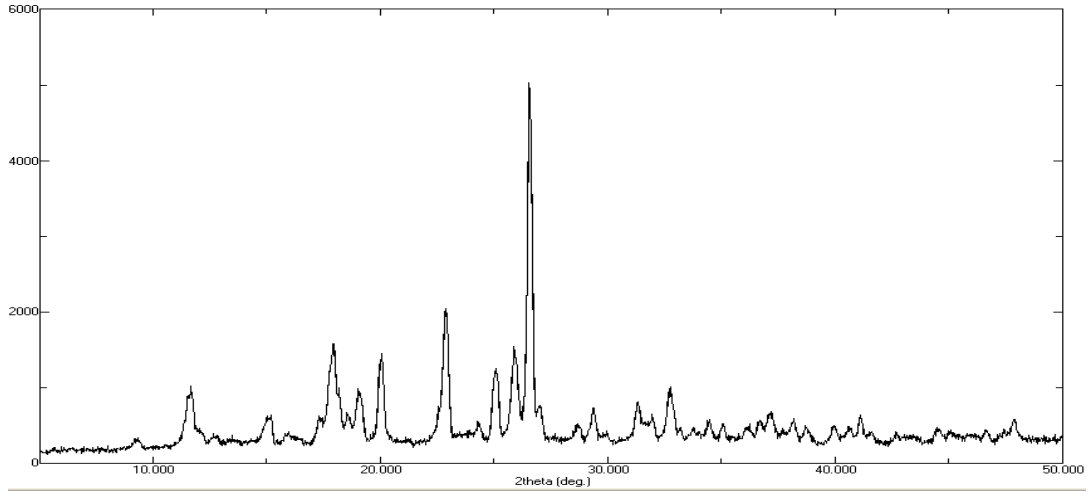


B

Fig. 1: DSC thermogram of (A) Ramipril and (B) Ramipril-β-cyclodextrin cocrystals.

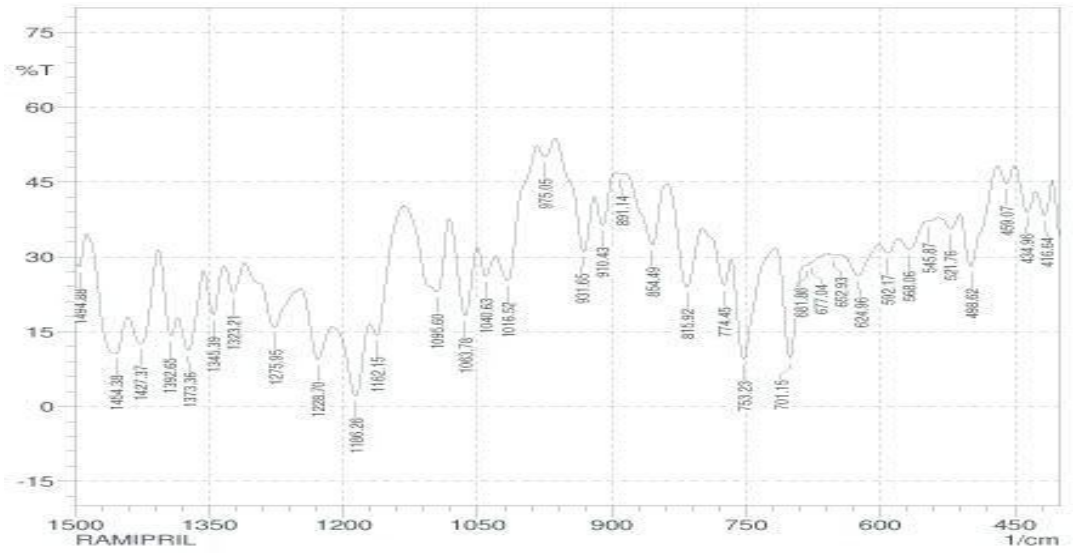


A

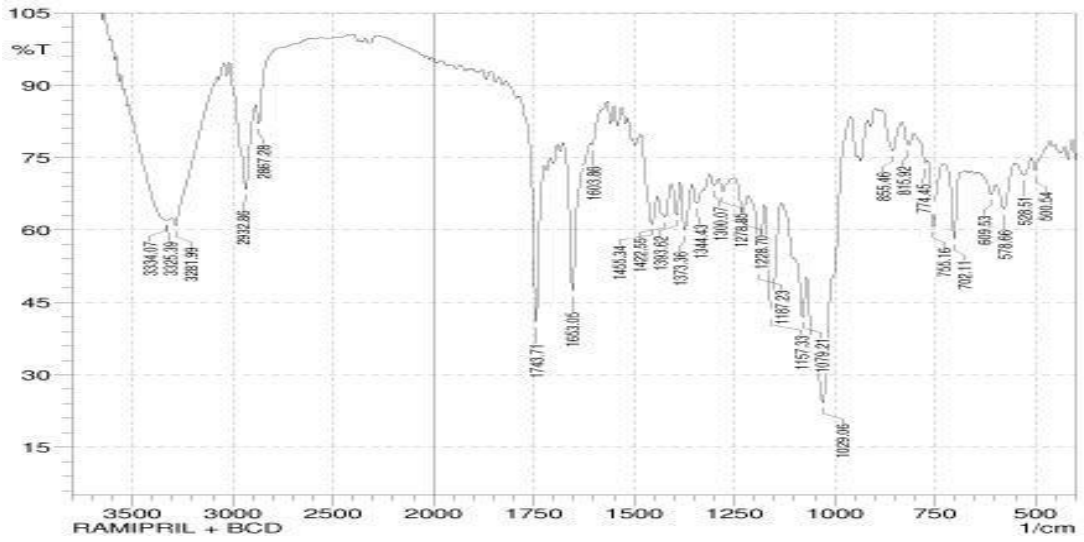


B

Fig. 2: Powder X-ray diffractogram of (A) Ramipril and (B) Ramipril-β-cyclodextrin cocrystals.

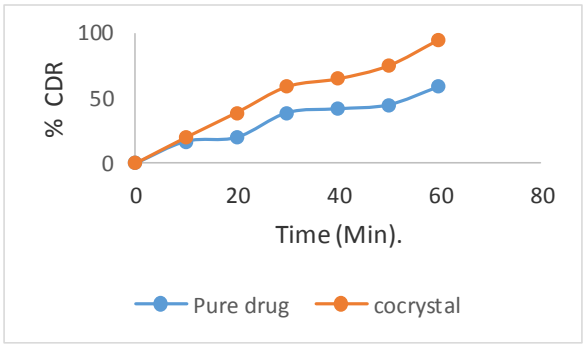


A



B

Fig. 3: Raman spectra of (A) Ramipril and (B) Ramipril-β-cyclodextrin cocrystals.



Cocrystal. The formation of hydrogen bonds between API and CCF was interpreted using Raman spectroscopy. FT-Raman spectra of ramipril and Ramipril-β-cyclodextrin cocrystals are shown in Fig. 3A and 3B respectively.

After the co-crystallization, the comparative micromeritic evaluation of ramipril and Ramipril-β-cyclodextrin cocrystals was performed. The results obtained showed that co-crystallization improves the flow and processing properties like the angle of repose, bulk density and tapped density. Carr's index and Hausner's ratio were improved as compared to pure drug.[28] The pre-formulation characteristics of drug and cocrystals are summarized in Table II.

Immediate release tablet dosage form of ramipril and Ramipril-β-cyclodextrin cocrystals was evaluated for different parameters like uniformity of weight, hardness, disintegration time and friability. All tablet evaluation parameters are given in Table III.[29,30]

The *in vitro* dissolution pattern of tablet formulation of ramipril and Ramipril-β-cyclodextrin cocrystals was studied in distilled water in (Fig. 4). Percent cumulative drug release was found to be 58.87±1.39 % and 92.65±1.21 % after 60 min, respectively, for Ramipril and Ramipril-β cyclodextrin cocrystal tablet, indicating the improved dissolution ramipril by crystal engineering.[31,32]

The results obtained from the stability study revealed that the tablets were stable for the complete duration of the study without any changes in physical parameters such as hardness and friability. The drug content was found to be 99.14±0.77 % after the study period which indicates that tablets were stable at 40 °C ± 2 °C and 75 % ± 5 % relative humidity. The stability analysis data is illustrated in Table IV33.

CONCLUSION

The development of bicomponent cocrystals improved the solubility of Ramipril by seven-fold with optimum stability. Novel crystalline form improved the physicochemical property of ramipril along with processability for the solid oral dosage form. Results from DSC, PXRD, and Raman spectroscopy also demonstrated the formation of bicomponent cocrystals. Solubility and in vitro dissolution studies showed a significant improvement in solubility and dissolution. These newly formed bicomponent cocrystals of aceclofenac are the better option for formulation development with efficient drug release than unprocessed aceclofenac.

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