Journal of Carcinogenesis
Covering Genesis to Therapeutics

Journal of Carcinogenesis, ISSN: 1477-3163
2025; Vol 24: Issue 9s, https://carcinogenesis.com/

Preparation and Evaluation of Pharmaceutical Cocrystals for Solubility Enhancement of Ketoconazole

Meena Singh[1*], Pankaj Masih[1], Naveen Gupta[1], Ganesh Prasad Patel[1], Brajmohan Kaushal[1]

[1]Department of Pharmacy, Madhyanchal Professional University, Bhopal, M. P.

Address for correspondence

Ms. Meena Singh

Research Scholar

Department of Pharmacy,

Madhyanchal Professional University Bhopal M.P.

ABSTRACT

Drug - drug co-crystallization is proposed as a new method to improve the solubility and hence the bioavailability of drugs. Cocrystal consists of two or more neutral molecular components in a crystal lattice with well- defined stoichiometric ratio. Formation of cocrystal mainly depends on functional group of APIs and co-formers to allow for the formation of hydrogen bonds or other weak type of interactions, mainly hydrogen bonding. The basis for co-formers selection is its ability to form non - covalent interactions especially hydrogen bonds with an APIs. The drug-drug cocrystals batches of ketoconazole and hydroquinone coformer were successfully prepared by various methods Co grinding, Solvent evaporation and Liquid assisted grinding. The physicochemical properties were characterized by various characterizations. Improved permeability of the drug is attributed to drug conformer interaction. This study showed the utility of the co-crystallization approach to improve bioavailability of drug after formulating tablet dosage form and various characterization and in vitro dissolution study was performed.

KEYWORDS: *Crystal engineering, hydroquinone coformers, ketoconazole, Dry Grinding, Solvent Evaporation, Liquid Assisted Grinding methods*

How to Cite: Meena Singh, Pankaj Masih, Naveen Gupta, Ganesh Prasad Patel, Brajmohan Kaushal., (2025) Preparation and Evaluation of Pharmaceutical Cocrystals for Solubility Enhancement of Ketoconazole, *Journal of Carcinogenesis, Vol.24, No.9s,* 526-532.

1. INTRODUCTION

Crystal engineering has been described as the ‘exploitation of noncovalent interactions between molecular or ionic components for the rational design of solid-state structures that might exhibit interesting electrical, magnetic, and optical properties [1]. It is also recognised that it ‘is becoming increasingly evident that the specificity, directionality, and predictability of intermolecular hydrogen bonds can be utilized to assemble supramolecular structures of, at the very least, controlled dimensionality. Supramolecular chemistry has grown around Lehn's analogy that ‘supermolecules are to molecules and the intermolecular bond, what molecules are to atoms and the covalent bond. If molecules are built by connecting atoms with covalent bonds, solid-state supermolecules (crystals) are built by connecting molecules with intermolecular interactions [2]. Modern crystal engineering initially began as a method for understanding the regioselectivity and product distribution in solid-state molecular reactions, termed topochemistry. This field has developed rapidly, particularly with the arrival of modern crystallographic techniques such as four circle diffractometers in the early 1970's followed by the introduction of area detector technology [3]. Crystal engineering now encompasses many aspects of solid-state intermolecular interactions, structure prediction, control and rationalisation, as well as the synthesis of novel molecular building blocks and crystalline materials, and may be broken down into the components of analysis and synthesis [4]. Within the notion of a crystal as a supramolecular entity lies certain key ideas central to the activity of crystal engineering. These are the nature of the crystallisation process at a molecular level, crystal packing, molecular interaction and directed molecular recognition, which will all be explored to some extent in this review and which should provide

some understanding of crystal engineering approaches as a means of addressing the challenges of low aqueous solubility [5]. Crystallisation is concerned with the evolution from solution or melt of the crystalline state. Within this area key issues include the formation of crystal nuclei, the influence of crystallisation conditions, and the overlap between the concepts of the growth unit, and an understanding of how the overall shape of a crystal evolves [6]. It is within the notion of the growth unit that a distinct link with the supramolecular concept of a synthon is achieved. The term 'synthon' was originally introduced to describe synthetic organic structural features [7]. Ketoconazole is widely recommended orally for the treatment of various fungal infections. One of the major disadvantages of oral therapy in the treatment of skin infections like Seborrhoeic dermatitis and Psoriasis is rapid relapse on the cessation of therapy and the risk of hepatotoxicity. Numerous approaches have been pursued to improve solubility and dissolution rate of poorly soluble drugs, such as salt preparation, solid dispersion, microemulsification, self-nanoemulsification, cosolvency, inclusion complex formation with cyclodextrin, choosing the right polymorphs, and nanoparticles formation. Cocrystals can be defined as molecular crystals that contain more than one of different molecules typically a drug and a cocrystal former "coformers" in the same structure of crystal lattice, which exists as solids at ambient conditions, bonded together by weak intermolecular interactions such as hydrogen bonding, van der waals forces, and π–π stacking. In pharmaceutical systems, the most popular bonds that form the foundation of molecular recognition phenomena are 'hydrogen bonds' and were responsible for the production of families of molecular networks in the crystalline molecules [8]. Cocrystallization involves the formation of a crystalline structure composed of the API and a co-former, typically a pharmaceutically acceptable compound that interacts with the API through non-covalent bonds, such as hydrogen bonds or van der Waals forces. Importantly, cocrystals do not alter the molecular identity of the API but modify its physicochemical properties by altering the drug's crystal lattice structure. This technology has already been explored in commercial formulations, demonstrating its potential to improve drug solubility and dissolution rates while maintaining the pharmacological integrity of the drug [9]. There is a critical need for an innovative formulation strategy to enhance ketoconazole's solubility and bioavailability, which could lead to improved therapeutic effectiveness and patient outcomes.

2. MATERIAL AND METHODS

Preparation of Cocrystal for solubility enhancement

Dry Grinding Method: Three formulas were prepared at different stoichiometric ratio (1:1, and 1:2) of ketoconazole and hydroquinone as a conformer as shown in table 1. Drug and coformer were mixed in a mortar using pestle and ground for 45 minutes to form cocrystals. These cocrystals have dried overnight at ambient temperature, then stored in tightly closed containers.

Solvent Evaporation Method: Ketoconazole with hydroquinone were carefully weighed at different stoichiometric ratio (1:1, and 1:2) as shown in table 1. Each compound was dissolved in ethanol separately. The two solutions were mixed and sonicated for a few minutes, and then the solution of both components was poured into a Petri dish. The prepared solution was allowed to evaporate at room temperature until the solution is completely dry. The obtained cocrystal solids were stored in a tightly closed container for further evaluation [10].

Liquid Assisted Grinding: Liquid assisted grinding technique was used to prepare cocrystals using hydroquinone as coformer. The formation of ketoconazole cocrystals was performed in the same molar ratio (1:1, and 1:2) as shown in table 1. Grinding of a mixture of drug and coformers was carried out in mortar and pestle for 30 minutes with the addition of 2.5 mL ethanol dropwise, and then the wet crystals were dried in an oven and stored for further analyses. In this technique, ethanol acts as a promoter, either as "media" that enables molecular diffusion or as an essential element that forms multi-components inclusion framework and has been used to enhance supramolecular selectivity in crystalline systems. It can be described the effect of the solvent as a catalytic, so that it is not part of the final product, because of its small amount used [11].

Table 1: Preparation of ketoconazole cocrystal with hydroquinone conformer

F. Code	Ketoconazole: Hydroquinone molar ratio	Cocrystallization methods
KHC1	01:01	Co grinding
KHC2	01:02	Co grinding
KHS1	01:01	Solvent evaporation
KHS2	01:02	Solvent evaporation
KHL1	01:01	Liquid assisted grinding
KHL2	01:02	Liquid assisted grinding

Characterization of cocrystals:

Saturation Solubility: An excess quantity of drugs and the formulated cocrystals was added to 10 mL vials containing distilled water to determine the saturation solubility. The vials were immersed in shaker water bath, subjected to agitation and allowed to stand for equilibration for 24 hours. Then, the Filtration and dilution of samples were done, and the concentration of drugs was determined from the absorbance measurement at 260 nm for ketoconazole by using an ultraviolet-visible light double beam spectrophotometer.

Drug Content: An accurate weight of cocrystal powder equivalent to 10 mg of pure drug was taken and dissolved in a 60 mL of 0.1 N HCl and the volume was completed to 100 mL in a volumetric flask. The resulting solution was filtered using Whatman filter paper 41 and the absorbance of the solution was measured at 260 nm for ketoconazole by using an ultraviolet light double beam spectrophotometer [12].

Infrared spectroscopy: Infrared spectroscopy was employed to determine the possible interaction between drug and co-former. The samples were dispersed in KBr pellet and scanned using PerkinElmer IR spectrophotometer between 4000 - 400 cm-1 with resolution of 4 cm-1.

Preparation of tablet dosage form: Equivalent to 10 mg content of drugs were prepared by mixing required quantities of Microcrystalline cellulose (Avicel PH - 102), Di-basic calcium phosphate dihydrate, lactose as filler, starch potato (internal binder) and PVP - K30 (10 % solution in iso-propyl alcohol) as a external binder. The wet granulated mass passed through a mesh # 10 and dried at 60 ºC for 1h in a hot air oven. The dried granules were sized by passing through a sieve # 14. The complete batch of dried granules was collected and mixed with talc as glidant and magnesium stearate as lubricating agent. The granules were compacted into tablets using single-punch tablet compression machine (Khera Instruments Pvt. Ltd., New Delhi), fitted with 8.0 mm flat-faced punches. Compression was controlled to produce a 5-kg tablet-crushing strength (Table 2) [13].

Table 2: Preparation of ketoconazole hydroquinone cocrystal tablet

Ingredients	Amount (mg /tablet)					
	KHC1T	KHC2T	KHS1T	KHS2T	KHL1T	KHL2T
Ketoconazole hydroquinone cocrystal	KHC1 (200mg)	KHC2 (200mg)	KHS1 (200mg)	IHS2 (200mg)	KHL1 (200mg)	KHL2 (200mg)
Microcrystalline cellulose (Avicel PH - 102)	140	140	140	140	140	140
Sodium starsh glycollate	15	15	15	15	15	15
Di-basic calcium phosphate dihydrate (DBP)	55	55	55	55	55	55
Lactose	50	50	50	50	50	50
Starch (Potato)	30	30	30	30	30	30
Talc (Purified)	5	5	5	5	5	5
Magnesium stearate	5	5	5	5	5	5
Polyvinyl pyrrolidone K-30	10 % w/v in iso-propyl alochol					
Total weight of tablet	500 mg					

Characterization of Tablet:

Flow properties of granules: The flow properties of drug powder were characterized in terms of Carr's index (%), Hausner's ratio and angle of repose (ɵ). The Carr's index ((I_C)) and Hausner's ratio (H_R) of drug powders were calculating according to previous discuss equations.

Weight variation: Not more than two of the individual weights deviate from the average weight by more than the percent shown below and none deviates by more than twice that percent [14].

Thickness: The thickness of tablets was performed on 20 tablets from each formulation. The Vernier caliper was used for the study.

Hardness: Hardness of tablet is defined as the force required to break a tablet a in a diametric direction. A tablet was placed between two anvils. Force was applied to anvils and crushing strength that causes the tablet to break was recorded. Hardness is thus the tablet crushing strength. Monsanto tester is used for hardness testing.

Friability: Weigh 10 tablets and place in a friabilator chamber rotated at 25 rpm and they are dropped on distance of 6 inches. The chamber is allowed to rotate for 100 revolutions. Then the tablets are removed, dusted and again the weight is taken. The difference in the weigh is calculated and the weight loss should not be more than 1%. [15-16]

In- vitro dissolution study:
In-vitro study was performed by using USP type II dissolution apparatus with rotation speed of set to 100 rpm. About 900 mL of 0.1N Hcl at 37 ± 0.5 °C was used as a dissolution media. At predetermined time intervals 1 mL samples were withdrawn, filtered through 0.45 µm membrane and 1 mL blank dissolution medium was added for replenishing of the dissolution medium. The amount of dissolved drug was determined at 260 nm for ketoconazole using a UV spectrophotometer [17].

3. RESULTS AND DISCUSSION

Saturation Solubility; The saturation solubility of pure ketoconazole in 0.1N HCl (pH 1) at 37°C was found to be 0.8 mg/mL after 24 hours. On the other hand, the cocrystal forms of ketoconazole with hydroquinone at the molar ratio 1:1 and 1:2 prepared by dry grinding method, solvent evaporation method and liquid assisted grinding showed an increase in drugs solubility after 24 hours under the same conditions as the pure drug. Cocrystals of ketoconazole prepared by solvent evaporation method at ratio 1:2 showed the highest improvement in dynamic solubility (1.6 mg/mL).

Table 3: Solubility of ketoconazole cocrystal with hydroquinone conformer in 0.1 N HCl (pH 1) at 37˚C and after 24 hours equilibration

F. Code	Ketoconazole: Hydroquinone molar ratio	Cocrystallization methods	Solubility (mg/ml)	Drug content (%)
KHC1	01:01	Co grinding	6.87	97.41
KHC2	01:02	Co grinding	7.14	98.04
KHS1	01:01	Solvent evaporation	9.88	98.41
KHS2	01:02	Solvent evaporation	10.98	98.78
KHL1	01:01	Liquid assisted grinding	8.01	96.46
KHL2	01:02	Liquid assisted grinding	8.87	97.01

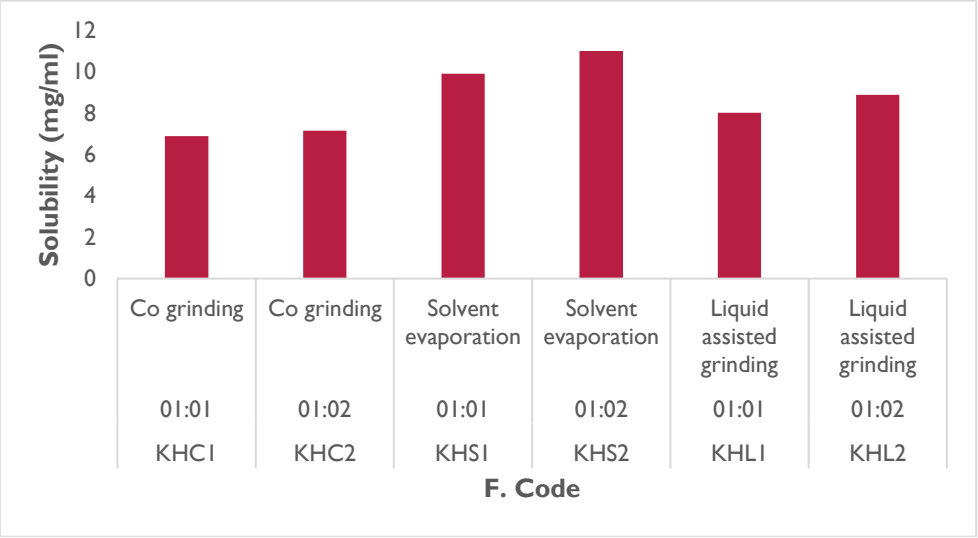

Figure 1: Solubility of ketoconazole cocrystal with hydroquinone conformer in 0.1 N HCl (pH 1) at 37˚C and after 24 hours equilibration

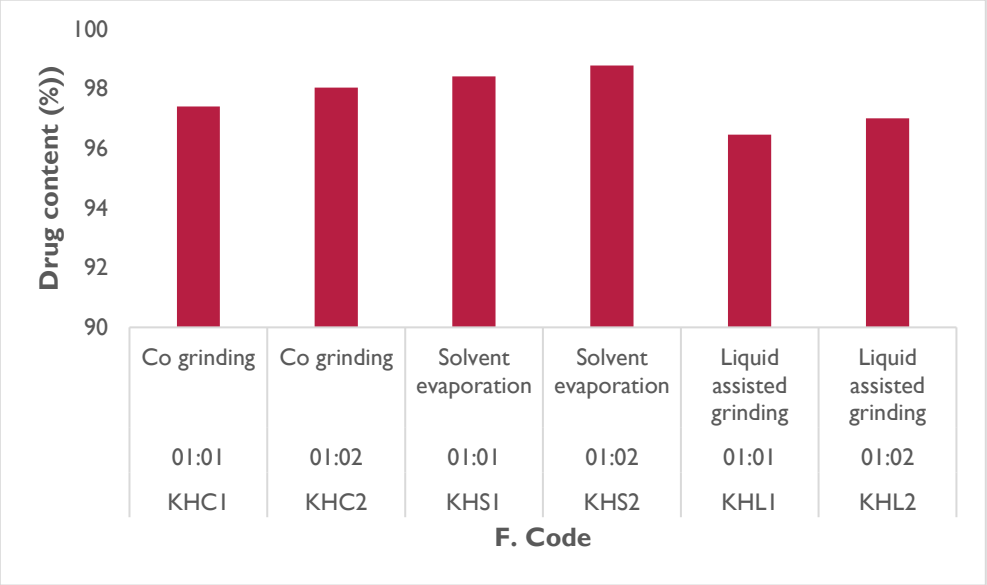

Figure 2: Drug content of ketoconazole cocrystal with hydroquinone conformer in 0.1 N HCl (pH 1) at 37˚C and after 24 hours equilibration

Fourier Transform-infrared (FTIR) Spectroscopy Analysis: Fourier transformation spectroscopies are commonly used as a tool for letting recognition of molecular modification in cocrystal lattice by hydrogen bond formation. The spectrum of mixture (Figure 3) of KRS2; Ketoconazole cocrystal with resorcinol conformer with Solvent evaporation method at a ratio of (1:2) shows the characteristic peaks for both compounds indicating there is no chemical interaction has occurred between them. The FTIR spectroscopy is used to confirm cocrystal formation because of its ability to detect differences in the chemical structures of samples indicative of the formation of hydrogen bonding. FTIR spectrum of the formulated cocrystals by solvent evaporation method showed a reduction in the intensity for lowering of frequency is the function of degree and strength of hydrogen bonding.

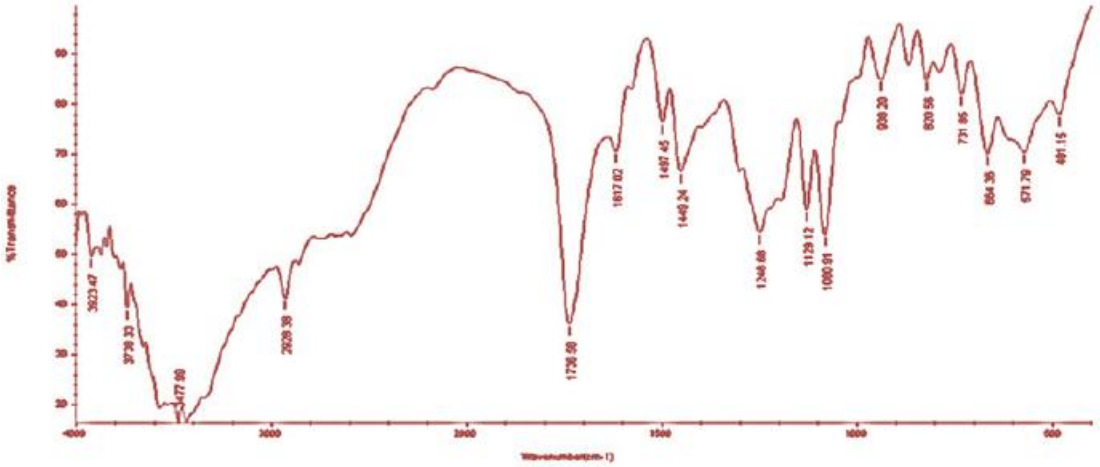

Figure 3: FTIR spectrum of KRS2; Ketoconazole cocrystal with resorcinol conformer with Solvent evaporation method

Characterization of tablet dosage form: The blend was initially characterized for pre-compression and post- compression parameters. Pre-compression characterization was done for angle of repose, bulk density, tapped density, Carr's index, and Hausner's ratio. Post-compression characterization includes thickness, hardness, friability, weight variation, drug content and in-vitro drug release. %. The average weights of the entire prepared tablet were 498.07±.03 mg to 509.14±.02 mg which was within the specified limit. The thickness of all the tablets was in the range of 2.48 to 2.52 um. The hardness of all the formulated tablets was found to be in the range of 5.94-6.01 kg/cm^2. Friability was found to be 0.76 to 0.93. The drug content of the entire prepared tablet was found to be 97.14 to 99.12 **(Table 4)**. From permeability study observations and benefits of ketoconazole, cocrystal batch prepared using in 1:2 hydroquinone ratio by Co grinding, Solvent evaporation

and liquid assisted grinding method was further selected for in vitro drug release study. To conduct in-vitro dissolution study, prepared cocrystals were developed into a tablet using direct compression method. These developed tablets showed hardness 5-6 kg/cm2, friability of 0.51 – 0.92%. In-vitro dissolution study was performed to compare release of both drugs from prepared cocrystals and release study is graphically presented KTC-RES % drug release tablet was found to be 70 % to 95 % for cocrystal tablet at the end 12 h (Figure 4).

Table 4: Pre-compression characterization of ketoconazole hydroquinone cocrystal tablet

Formulation code	Bulk density (g/cm^3)	Tapped density (g/cm^3)	Carr's index (%)	Hausner's Ratio	Angle of Repose (Θ)	Weight variation (mg)	Thickness (mm)	Diameter (mm)	Hardness (kg/cm^2)	Friability (%)	Drug content (%)
KHC1T	0.347	0.425	22.22	1.12	23.18	507.09±0.02	2.48±0.02	8.1±0.01	6.01±0.12	0.87±0.39	98.25±1.31
KHC2T	0.328	0.436	22.35	1.15	23.15	504.01±0.02	2.52±0.02	7.8±0.01	5.98±0.14	0.76±0.26	99.12±1.24
KHS1T	0.312	0.424	23.15	1.12	24.12	503.08±0.01	2.51±0.01	8.0±0.03	5.94±0.23	0.79±0.34	98.14±1.16
KHS2T	0.321	0.418	22.06	1.16	25.06	498.07±0.03	2.48±0.02	7.8±0.02	6.04±0.14	0.93±0.37	97.14±1.35
KHL1T	0.322	0.429	23.34	1.17	24.24	502.13±0.05	2.52±0.01	8.0±0.03	5.98±0.12	0.84±0.36	98.14±1.24
KHL2T	0.318	0.423	23.16	1.13	23.98	509.14±0.02	2.48±0.02	8.1±0.03	6.01±0.16	0.86±0.46	97.16±1.21

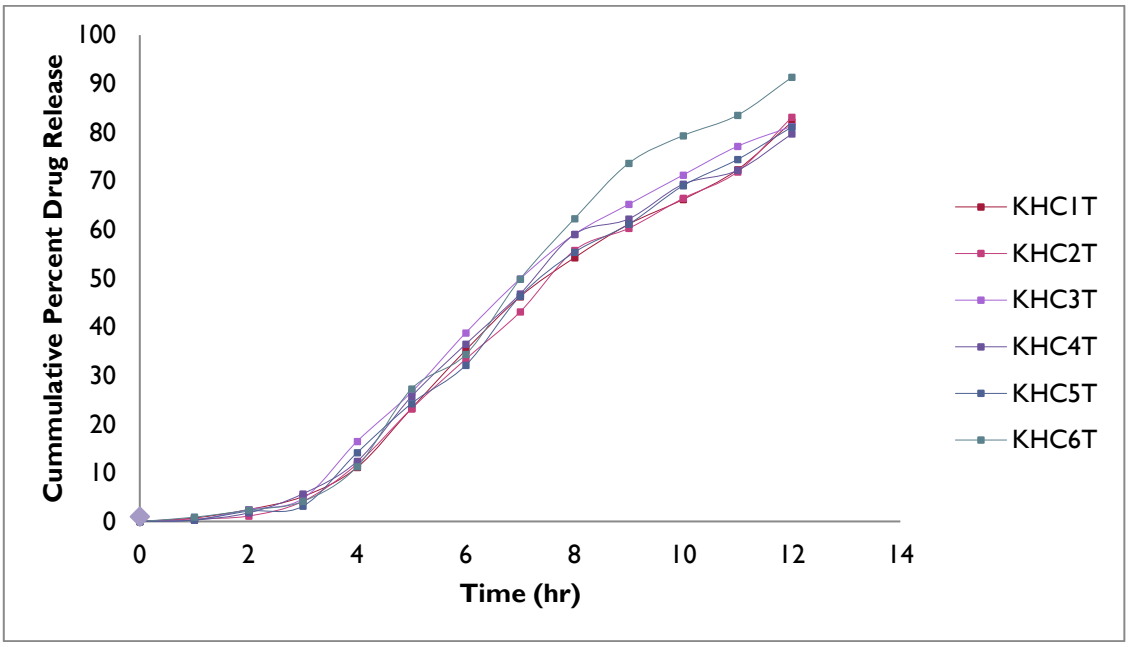

Figure 4: Zero-order release of ketoconazole hydroquinone cocrystal tablet (KHC1T – KHC6T)

Summary and Conclusion: The future for co-crystals appears bright, but there are problems which still have to be solved. The solubility, stability and bioavailability of drugs could be improved by the use of co-crystals in the field of pharmaceutics. The formation of new classes of pharmaceutical entities can result from the use of co-crystals which have a higher activity and a decreased activity. The use of co-crystals also will increase the Intellectual property protection of the drug molecule which is an elegant way of preserving patentability for an extended period. The improvement in the physical and chemical properties, enables the extension of the application of co-crystals to new fields of materials reserch like energy storage, catalysis and electronics. There is also the need for the implementation of the regulatory framework

concerning pharmaceutical co-crystals so that appropriate review and approval systems are in place. The composition in relation to geometry and strength of interactions leading to co-crystals and what maintains their integrity is also being investigated by scientists. Systems of design and characterization of co-crystals on the basis of their intrinsic properties will possibly be possible through the use of predictive techniques and computational methods. There is great promise for co-crystals in a number of fields including pharmaceuticals and materials research. The problems of synthesis, realization, regulatory attainment and methods of operation must be solved before co-crystals achieve their maximum potentiality. The necessary interaction between scientists, engineers and regulatory bodies points towards a bright future for co-crystals.

REFERENCES

[1] Chettri A, Subba A, Singh GP, Bag PP. Pharmaceutical co-crystals: A green way to enhance drug stability and solubility for improved therapeutic efficacy. Journal of Pharmacy and Pharmacology. 2024 Jan 1;76(1):1-2.

[2] Wostatek T, Chirala VR, Stoddard N, Civas EN, Pimputkar S, Schimmel S. Ammonothermal crystal growth of functional nitrides for semiconductor devices: Status and potential. Materials. 2024 Jun 25;17(13):3104.

[3] Wang Z, Li S, Li Q, Wang W, Liu M, Yang S, Zhang L, Yang D, Du G, Lu Y. A novel cocrystal of daidzein with piperazine to optimize the solubility, permeability and bioavailability of daidzein. Molecules. 2024 Apr 10;29(8):1710.

[4] Soopy AK, Liu S, Najar A. Enhancement of Photodetector Characteristics by Zn-Porphyrin-Passivated MAPbBr3 Single Crystals. Nanomaterials. 2024 Jun 21;14(13):1068.

[5] Megantara S, Rusdin A, Budiman A, Shamsuddin S, Mohtar N, Muchtaridi M. Revolutionizing antiviral therapeutics: unveiling innovative approaches for enhanced drug efficacy. International Journal of Nanomedicine. 2024 Dec 31:2889-915.

[6] Ishtiaq M, Manzoor H, Khan IU, Asghar S, Irfan M, Albekairi NA, Alshammari A, Alqahtani AF, Alotaibi S, Munir R, Shah PA. Curcumin-loaded soluplus® based ternary solid dispersions with enhanced solubility, dissolution and antibacterial, antioxidant, anti-inflammatory activities. Heliyon. 2024 Jul 30;10(14).

[7] Ye S, Chen T, Yu J, Wang S, Li S, Wang J, Fu Y, Zhu Y, Wang M, Lu X, Ma Z. Enhanced crystal network and charge transfer of non-fused ring electron acceptors via interchain interaction for efficient and stable organic solar cells. Energy & Environmental Science. 2024;17(14):5137-46.

[8] Kumari L, Choudhari Y, Patel P, Gupta GD, Singh D, Rosenholm JM, Bansal KK, Kurmi BD. Advancement in solubilization approaches: A step towards bioavailability enhancement of poorly soluble drugs. Life. 2023 Apr 27;13(5):1099.

[9] Budiman A, Rusdin A, Aulifa DL. Current techniques of water solubility improvement for antioxidant compounds and their correlation with its activity: molecular pharmaceutics. Antioxidants. 2023 Feb 4;12(2):378.

[10] Wang S, Wang T, Zhang S, Dong Z, Chevali VS, Yang Y, Wang G, Wang H. Enhancing fiber-matrix interface in carbon fiber/poly ether ether ketone (CF/PEEK) composites by carbon nanotube reinforcement of crystalline PEEK sizing. Composites Part B: Engineering. 2023 Feb 15;251:110470.

[11] Xia M, Jiang Y, Cheng Y, Dai W, Rong X, Zhu B, Mei X. Rucaparib cocrystal: Improved solubility and bioavailability over camsylate. International Journal of Pharmaceutics. 2023 Jan 25;631:122461.

[12] Song X, Luo Y, Zhao W, Liu S, Wang Y, Zhang H. Preparation and characterization of lutein co-amorphous formulation with enhanced solubility and dissolution. Foods. 2024 Jun 26;13(13):2029.

[13] Liu R, Wang L, Yu X, Xu Z, Gong H, Zhao T, Hu F, Shen B. Magnetocrystalline anisotropy study of Co-substituted M-type strontium hexaferrite single crystals. Ceramics International. 2023 Jan 15;49(2):1888-95.

[14] Bhalani DV, Nutan B, Kumar A, Singh Chandel AK. Bioavailability enhancement techniques for poorly aqueous soluble drugs and therapeutics. Biomedicines. 2022 Aug 23;10(9):2055.

[15] Bolla G, Sarma B, Nangia AK. Crystal engineering of pharmaceutical cocrystals in the discovery and development of improved drugs. Chemical reviews. 2022 Jun 1;122(13):11514-603.

[16] Choi MJ, Woo MR, Choi HG, Jin SG. Effects of polymers on the drug solubility and dissolution enhancement of poorly water-soluble rivaroxaban. International Journal of Molecular Sciences. 2022 Aug 22;23(16):9491.

[17] Li B, Hu Y, Wu T, Feng Y, Jiang C, Du H, Lu S. Apigenin-oxymatrine binary co-amorphous mixture: Enhanced solubility, bioavailability, and anti-inflammatory effect. Food Chemistry. 2022 Mar 30;373:131485.