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Physical stability enhancement of theophylline via cocrystallization

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Abstract

The crystal form adopted by the respiratory drug theophylline was modified using a crystal engineering strategy in order to search for a solid material with improved physical stability. Cocrystals, also referred to as crystalline molecular complexes, were prepared with theophylline and one of several dicarboxylic acids. Four cocrystals of theophylline are reported, one each with oxalic, malonic, maleic and glutaric acids. Crystal structures were obtained for each cocrystal material, allowing an examination of the hydrogen bonding and crystal packing features. The cocrystal design scheme was partly based upon a series of recently reported cocrystals of the molecular analogue, caffeine, and comparisons in packing features are drawn between the two cocrystal series. The theophylline cocrystals were subjected to relative humidity challenges in order to assess their stability in relation to crystalline theophylline anhydrate and the equivalent caffeine cocrystals. None of the cocrystals in this study converted into a hydrated cocrystal upon storage at high relative humidity. Furthermore, the theophylline:oxalic acid cocrystal demonstrated superior humidity stability to theophylline anhydrate under the conditions examined, while the other cocrystals appeared to offer comparable stability to that of theophylline anhydrate. The results demonstrate the feasibility of pharmaceutical cocrystal design based upon the crystallization preferences of a molecular analogue, and furthermore show that avoidance of hydrate formation and improvement in physical stability is possible via pharmaceutical cocrystallization.

Keywords: Theophylline; Cocrystal; Crystalline molecular complex; Crystal engineering; Stability; Hydrogen bond; Hydrate; Relative humidity

1. Introduction

Crystal engineering has been investigated recently as a means of tailoring the physicochemical properties of an active pharmaceutical ingredient (API) (Oswald et al., 2002; Bailey Walsh et al., 2003; Fleischman et al., 2003; Almarsson and Zaworotko, 2004; Childs et al., 2004; Trask et al., 2005). The formation of pharmaceutical cocrystals (crystalline molecular complexes) involves the incorporation of a given API with another pharmaceutically acceptable molecule in the crystal lattice. The resulting multi-component crystal form will possess a distinct physicochemical profile, potentially enabling improvements in properties such as solubility, melting point or physical stability.

The formation of salts, or crystalline ionic complexes, is a well-established means of altering the physicochemical properties of an API (Stahl and Wermuth, 2002a), but possesses several inherent drawbacks. Salt formation requires at least one ionizable center on the API of interest. As a result, non-ionizable pharmaceutical molecules are incapable of salt formation, and thus may be considered to be at a greater risk in terms of their pharmaceutical profiles. An additional limitation to salt formation is that the number of non-toxic, pharmaceutically acceptable acids and bases that may be implemented in salt formation is relatively small. Despite the prevalence of salt formation in the pharmaceutical industry, one survey revealed that there are only 10 salt-forming acidic counter-ions with a market usage rate of over 1%, and the number of comparable basic counter-ions is even fewer (Bighley et al., 1996).

Pharmaceutical cocrystallization, which has only recently gained widespread attention as a means of modifying the physicochemical properties of APIs (Almarsson and Zaworotko, 2004), has at least two inherent advantages over salt formation. First, because cocrystal formation involves the complexation of neutral molecules as opposed to ions, pharmaceutical cocrystallization may potentially be employed with all APIs, including acidic, basic and non-ionizable molecules. Second, there

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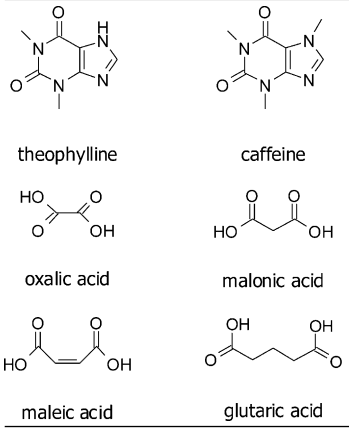
Table 1
Chemical structures of molecules under consideration in the present study



is a large number of potential 'counter-molecules' which may be considered to be non-toxic, possibly increasing the scope of pharmaceutical cocrystallization over salt formation; such substances may include food additives, preservatives, pharmaceutical excipients, vitamins, minerals, amino acids and other biomolecules, as well as other APIs.

Due to the large number of counter-molecules available for possible cocrystallization, a rational approach to cocrystal design is required to maximize experimental efficiency. Two important aspects of cocrystallization experiment design include evaluating the robustness of potential intermolecular interactions (i.e., assessing the likelihood of formation of specific interactions, such as hydrogen bond motifs) and considering general hydrogen bonding rules. The evaluation of intermolecular interaction robustness may be performed by analyzing trends within the Cambridge Structural Database (CSD) (Allen et al., 1999; Allen, 2002) as a whole, or by applying information from previous crystal engineering studies which have demonstrated the robustness of certain specific intermolecular interactions (or, ideally, by combining both of these approaches).

Hydrogen bonds are often employed in cocrystal design due to their inherently robust and directional nature. In cases when hydrogen bonding is expected to play a role in cocrystal formation (e.g. when good hydrogen bond donors and acceptors are available) it is of importance to consider general hydrogen bonding rules (Etter, 1990). From a number of systematic studies of cocrystals it was recognized that, in general, all good hydrogen bond donors and acceptors would be used in hydrogen bonding. Furthermore, of particular importance to the design of cocrystals, it was noted that the best hydrogen bond donor tends to interact with the best hydrogen bond acceptor in a given crystal structure. This 'best-donor–best-acceptor' rule can be of great utility in the design of specific hydrogen bonding interactions.

The API under consideration in the current report is theophylline (Table 1), a drug of use in the treatment of respiratory diseases such as asthma. From a physicochemical standpoint, theophylline represents a challenge to formulators in that it is known to interconvert between crystalline anhydrate and monohydrate forms as a function of relative humidity (RH). The possibility of crystalline hydrate formation complicates the design of a consistent, reproducible formulation process for an API in the drug development process (Khankari and Grant, 1995). Reversible hydrate formation is particularly problematic, as it indicates that neither the anhydrate nor the hydrate may be fully stable across a range of common processing conditions. The interest in theophylline as a model API for studying hydrate/anhydrate interconversion is evidenced by the number of reports involving its hydration behavior (Shefter et al., 1973; Herman et al., 1988; Otsuka and Kaneniwa, 1988; Suzuki et al., 1989; Puttipipatkhachorn et al., 1990; Rodríguez-Hornedo et al., 1992; Agbada and York, 1994; Duddu et al., 1995; Zhu et al., 1996; Phadnis and Suryanarayanan, 1997; Suihko et al., 1997; Ticehurst et al., 2002). The crystal structures of a number of theophylline cocrystals have been reported to date [CSD reference codes CSATEO (with 5-chlorosalicylic acid); DUXZAX (with urea); SULTHE (with sulfathiazole); THOPBA (with phenobarbital); TOPPNP (with *p*-nitrophenol); WUYROX (with *N*-(2-aminoethyl)-carbamate); ZAYLOA (with 5-fluorouracil and water); ZEXTIF (with *p*-nitroaniline)], but a systematic crystal engineering study aimed at improving the physical stability of theophylline has not, to our knowledge, been reported.

A chemical analogue of theophylline, the model API caffeine (Table 1) also exhibits reversible crystalline hydrate formation as a function of RH (Griesser and Burger, 1995; Edwards et al., 1997). We recently reported a crystal engineering study with caffeine in which one of several new caffeine cocrystals was found to be physically stable across all relative humidity conditions (Trask et al., 2005). Similarly, in the current report, the goal of the study was to design and prepare a cocrystal of theophylline that exhibited enhanced physical stability as a function of RH. The design of the theophylline cocrystals was based upon the previously demonstrated cocrystallization preferences of caffeine, and therefore represented an evaluation of the use of cocrystal design information gleaned from cocrystals of close chemical variants: this is a topic of particular interest in the area of pharmaceuticals, where new molecular entities are often developed in groups of chemically similar analogues.

Despite the chemical similarity between the two molecules, cocrystal design involving theophylline represented an increase in complexity over that involving caffeine. The hydrogen bonding capability of theophylline includes the several hydrogen bond acceptors it shares with caffeine (two carbonyl oxygens and one basic nitrogen), but in addition, because it has one less methyl group than caffeine, theophylline also possesses a good N–H hydrogen bond donor. As evidence, theophylline is both weakly acidic and weakly basic, with corresponding pK_a and pK_b values of 8.6 and 11.5, respectively (Cohen, 1975). The presence of both donors and acceptors on theophylline adds an extra element of complexity in considering the design of hydrogen-bonded cocrystals of theophylline.

The presence of a hydrogen bond donor in theophylline is observed to play a significant role in the crystal packing of both the anhydrate and monohydrate crystal forms previously reported (Fig. 1a and b; CSD reference codes BAPLOT01 (Ebisuzaki et al., 1997) and THEOPH01 (Sun et al., 2002), respectively). Of particular interest is that the two crystal structures illustrate the 'best-donor–best-acceptor' hydrogen bond rule mentioned above. In the structure of theophylline anhydrate,

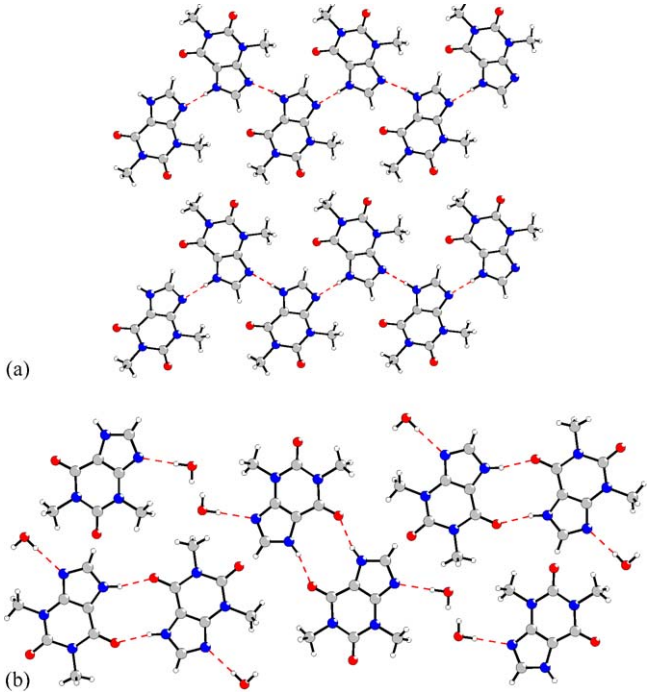


Fig. 1. Packing diagram of: (a) theophylline anhydrate showing hydrogen-bonded ribbons (Ebisuzaki et al., 1997) and (b) theophylline monohydrate, showing theophylline dimers (Sun et al., 2002).

the only good donor (the N–H) hydrogen bonds to the acceptor which would be expected to be strongest (the basic nitrogen). The resulting structure consists of chains of theophylline molecules linked by N–H···N hydrogen bonds (Fig. 1a). The crystal structure of theophylline monohydrate illustrates the effect of introducing a competing hydrogen bond donor: in the presence of water, the basic nitrogen of theophylline preferentially forms O–H···N hydrogen bonds. The N–H donor, adhering to the rule that all good donors and acceptors are typically used in hydrogen bonding, pairs with a carbonyl oxygen in an adjacent theophylline molecule to form a secondary N–H···O hydrogen bond. This secondary N–H···O interaction allows the theophylline molecules to form hydrogen-bonded dimers in a cyclic motif (Fig. 1b) that may be described in graph set notation (Etter et al., 1990) as R_2^2 (10) (the symbolism indicating that the motif takes the form of a hydrogen-bonded ring consisting of 10 atoms in total, 2 of which act as hydrogen-bond donors and 2 as hydrogen-bond acceptors).

By comparison of the theophylline anhydrate and monohydrate structures, and in the context of the aforementioned hydrogen bond rules, the R_2^2 (10) dimer motif is apparently favored only in the presence of a competing strong hydrogen bond donor. This hydrogen bond preference is confirmed by the only unionized crystalline complex in the CSD that involves theophylline and a carboxyl-containing counter-molecule, that of theophylline:5-chlorosalicylic acid, CSD reference code CSATEO (Shefter, 1969). These observations regarding the hydrogen-bonding behavior of theophylline in the presence of a strong proton donor have direct relevance to the present crystal engineering strategy.

The design strategy employed in the preparation of theophylline cocrystals involved cocrystallizing theophylline with a number of dicarboxylic acid counter-molecules with varying degrees of precedence as pharmaceutical counter-ions (Stahl and Wermuth, 2002b). The counter-molecules employed (and their aqueous pK_{a_1} and pK_{a_2} values; Stahl and Wermuth, 2002b) included oxalic acid (1.3, 4.3), malonic acid (2.8, 5.7), maleic acid (1.9, 6.2) and glutaric acid (4.3 and 5.2; see Table 1). Based upon the previously demonstrated preference for O–H···N bonds in the structure of theophylline hydrate, the caffeine:dicarboyxlic acid cocrystal series (Trask et al., 2005), and the previously mentioned CSD structure CSATEO, it was anticipated that any theophylline:dicarboxylic acid cocrystals obtained in this study would exhibit the same hydrogen bond arrangement. Also, as demonstrated with the caffeine cocrystal series, the dual approaches of solution crystallization and solid-state grinding (Trask and Jones, 2005) were employed to achieve theophylline cocrystal formation.

2. Experimental

2.1. Materials

Theophylline (minimum 99% chemical purity) was obtained from Fluka Chemie GmbH (Switzerland). All other chemical reagents were obtained from Sigma–Aldrich Co. Ltd. (Gillingham, UK) with a minimum chemical purity of 98%. Chemicals were used as received without further purification.

2.1.1. Theophylline

Powder XRD analysis of this material agreed with the PXRD pattern simulated from the known crystal structure of theophylline anhydrate, CSD reference code BAPLOT01 (Ebisuzaki et al., 1997).

2.1.2. Cocrystal A (2:1 theophylline:oxalic acid)

Cocrystal A material could be prepared by solution precipitation and by solid-state grinding. Material for RH stability evaluation was prepared by dissolving theophylline (2.047 g; 11.36 mmol) and oxalic acid (512 mg; 0.5 eq) in 40 ml chloroform and 11 ml methanol at reflux (55 °C). The solution was removed from heat and allowed to cool to ambient temperature. Solids then precipitated and were filtered. A single crystal for crystal structure analysis was obtained by slow evaporation from chloroform:methanol (ca. 20:1). Cocrystal A material was also prepared by grinding theophylline (470 mg; 2.61 mmol) with oxalic acid (118 mg; 0.5 eq) under the grinding conditions described in Section 2.2. The PXRD pattern of the material from grinding matched that simulated from the single crystal structure obtained from solution crystallization.

2.1.3. Cocrystal B (1:1 theophylline:malonic acid)

Cocrystal B material for RH evaluation was prepared by dissolving theophylline (587 mg, 3.26 mmol) and malonic acid (339 mg, 1.0 eq) in 40 ml chloroform and 2 ml methanol at reflux. The solution was removed from heat and seeded with phase-pure B obtained by grinding together theophylline (149 mg,

0.83 mmol) and malonic acid (86 mg, 1.0 eq). The stirred solution clouded as crystallization commenced, and the solvent was permitted to evaporate until the total volume was ca. 15 ml, at which time the slurry was filtered. The filtrate was allowed to evaporate slowly in order to yield single crystals for structure determination.

2.1.4. Cocrystal C (1:1 theophylline:maleic acid)

Cocrystal C material for RH evaluation was prepared by dissolving theophylline (2.02 g, 11.20 mmol) and maleic acid (1.30 g, 1.0 eq) in 35 ml of a 6:1 chloroform:methanol solvent mixture at reflux. The solution was removed from heat and allowed to cool while stirring continuously. Precipitation occurred and the resulting solids were vacuum filtered. While these solids (439 mg) contained excess theophylline anhydrate, the subsequent precipitate (478 mg) from the filtrate was found to be essentially phase-pure C. A single crystal for structure determination was harvested from a slowly evaporated acetonitrile solution of theophylline and excess maleic acid. A grinding experiment involving theophylline (421 mg, 2.37 mmol) and maleic acid (270 mg, 1.0 eq) revealed evidence of cocrystal C formation in the presence of unreacted starting materials.

2.1.5. Cocrystal D (1:1 theophylline:glutaric acid)

Cocrystal D material for RH evaluation was prepared by dissolving theophylline (496 mg, 2.75 mmol) and glutaric acid (368 mg, 1.1 eq) in 35 ml chloroform at reflux. The solution was removed from heat and seeded with nearly phase-pure cocrystal D material obtained by grinding theophylline (148 mg, 0.82 mmol) with glutaric acid (109 mg, 1.0 eq). The seeded solution was evaporated to ca. 12 ml total volume, and the precipitate was filtered. A single crystal was obtained from slow evaporation of an aliquot from the filtrate.

2.2. General methods

Relative humidity conditions were achieved at ambient temperature (ca. 20 °C) within sealed glass desiccator jars containing P_2O_5 for the 0% RH condition, distilled water for the 100% RH condition, and an appropriate saturated aqueous salt solution for other RH values: K_2CO_3 for 43%; NaCl for 75%; K_2SO_4 for 98% (Greenspan, 1977). Relative humidity conditions were monitored with humidity-indicator cards (Sigma–Aldrich Co. Ltd.).

In order to compare the stability of theophylline anhydrate to that of cocrystals A–D, samples of each were evaluated for physical stability at the conditions of 0%, 43%, 75% and 98% RH for time periods of 1 day, 3 days, 1 week and 7 weeks. Open glass vials containing 20–30 mg of powder were stored in the RH chambers at ambient temperature. The range of particle sizes of these powders was not strictly controlled. A vial was removed for each cocrystal material at each time point. Upon removal from the chamber, the samples were promptly evaluated for any form change by PXRD.

Solid-state grinding was performed with a Retsch MM200 Mixer Mill equipped with stainless steel 10 ml grinding jars and two 7-mm stainless steel grinding balls per jar. All grinding was performed for 60 min at a rate of 30 Hz. The temperature of the grinding jars following grinding experiments did not exceed ca. 30 °C.

Powder X-ray diffraction (PXRD) data was collected on a Philips X'Pert Pro diffractometer, using Ni-filtered Cu Kα radiation (λ = 1.5418 Å) at 40 kV and 40 mA with a X'Celerator RTMS detector. Each sample was analyzed between 4 and 40°2θ with a step size of ca. 0.02°2θ and a total scan time of 3 min 5 s. Powder XRD patterns were simulated from single crystal data using the program *MERCURY* (Bruno et al., 2002). Experimental PXRD patterns were compared to PXRD patterns simulated from the crystal structures of cocrystals A–D to confirm the composition of those materials.

Single crystal X-ray diffraction data was collected at 180 (2) K with a Nonius Kappa CCD diffractometer using Mo Kα radiation (λ = 0.71073 Å) and equipped with an Oxford Cryosystems cryostream. Data reduction and cell refinement were performed with the programs *DENZO* (Otwinowski and Minor, 1997) and *COLLECT* (Nonius, 1998). Multi-scan absorption corrections were applied with the program *SORTAV* (Blessing, 1995). Structures were solved by direct methods using *SHELXS*-97 (Sheldrick, 1997b) and refined on F^2 against all data using *SHELXL*-97 (Sheldrick, 1997a). All non-hydrogen atoms were refined with anisotropic displacement parameters. The OH and NH hydrogen atoms were located in different Fourier maps and refined isotropically. All other hydrogen atoms were placed geometrically and were allowed to ride during subsequent refinement. Experimental details of the structure determinations of cocrystals A–D are given in Table 2, and CIFs have been deposited with the Cambridge Crystallographic Data Centre (CCDC 283494–283497 contains the supplementary crystallographic data for this paper. These data can be obtained free of charge via www.ccdc.cam.ac.uk/data_request/cif, or by emailing data_request@ccdc.cam.ac.uk, or by contacting The Cambridge Crystallographic Data Centre, 12 Union Road, Cambridge CB2 1EZ, UK; fax: +44 1223 336033).

3. Results and discussion

3.1. Crystal packing features of theophylline cocrystals

Single crystal structures for cocrystals A–D are summarized in Table 2, and details of their packing features are described in turn below.

3.1.1. Cocrystal A (2:1 theophylline:oxalic acid)

Cocrystal A is a crystalline complex of theophylline with oxalic acid; oxalic acid is recognized as an acceptable pharmaceutical salt-forming acid with a limited precedence on the market (Stahl and Wermuth, 2002b). A 2:1 theophylline:oxalic acid stoichiometry was confirmed by crystal structure analysis on the resulting cocrystal (Fig. 2a). As was the case with every cocrystal obtained in this study, the acidic proton was located on the acid via single crystal XRD, thereby confirming the non-ionic character of the complex.

In the crystal structure of cocrystal A, each oxalic acid links two theophylline molecules; the intermolecular O–H···N

Table 2
Crystallographic data for theophylline cocrystals A–D

	Cocrystal A theophylline: oxalic acid (2:1)	Cocrystal B theophylline: malonic acid (1:1)	Cocrystal C theophylline: maleic acid (1:1)	Cocrystal D theophylline: glutaric acid (1:1)
Experimental formula	$2(C_7H_8N_4O_2)\cdot C_2H_2O_4$	$C_7H_8N_4O_2\cdot C_3H_4O_4$	$C_7H_8N_4O_2\cdot C_4H_4O_4$	$C_7H_8N_4O_2\cdot C_5H_8O_4$
Formula weight	450.38	284.24	296.25	312.29
Crystal system	Monoclinic	Monoclinic	Triclinic	Monoclinic
Space group	$P2_1/c$	$C2/c$	P-1	$P2_1/c$
a (Å)	5.8205 (12)	17.1882 (4)	7.9707 (16)	9.5994 (2)
b (Å)	16.609 (3)	8.3879 (2)	8.6133 (17)	19.8971 (4)
c (Å)	9.806 (2)	17.6293 (4)	10.665 (2)	15.3256 (4)
α (°)	90	90	69.55 (3)	90
β (°)	99.83 (3)	105.6837 (17)	72.53 (3)	107.885 (1)
γ (°)	90	90	71.24 (3)	90
V ($Å^3$)	934.1 (3)	2447.03 (10)	634.9 (2)	2785.74 (11)
Z	2	8	2	8
θ Range (°)	3.55–27.51	3.84–27.49	3.76–24.91	3.61–27.50
Data/restraints/parameters	2137/0/153	2795/0/195	2167/0/238	6358/0/425
ρ_{calc} (g cm^{-3})	1.601	1.543	1.550	1.489
T (K)	180 (2)	180 (2)	180 (2)	180 (2)
$R1$	0.0457	0.0395	0.0564	0.0523
w$R2$	0.1138	0.1020	0.1161	0.1174

hydrogen bonds between the acid and each theophylline represent the fulfillment of the 'best-donor–best-acceptor' rule, whereby the acid and the basic nitrogen play these respective roles. Furthermore, akin to the theophylline hydrate crystal structure, secondary N–H···O hydrogen bonds permit theophylline dimer formation.

The crystal packing of cocrystal A is similar in some respects to the reported cocrystal of caffeine with oxalic acid (Fig. 2b). Both crystal structures demonstrate a 2:1 drug:acid ratio, with the oxalic acid spanning between two drug molecules via hydrogen bonds. Furthermore, the anticipated O–H···N bond is observed in each structure, with the equivalent nitrogen on each drug molecule hydrogen bonding to the carboxylic acid.

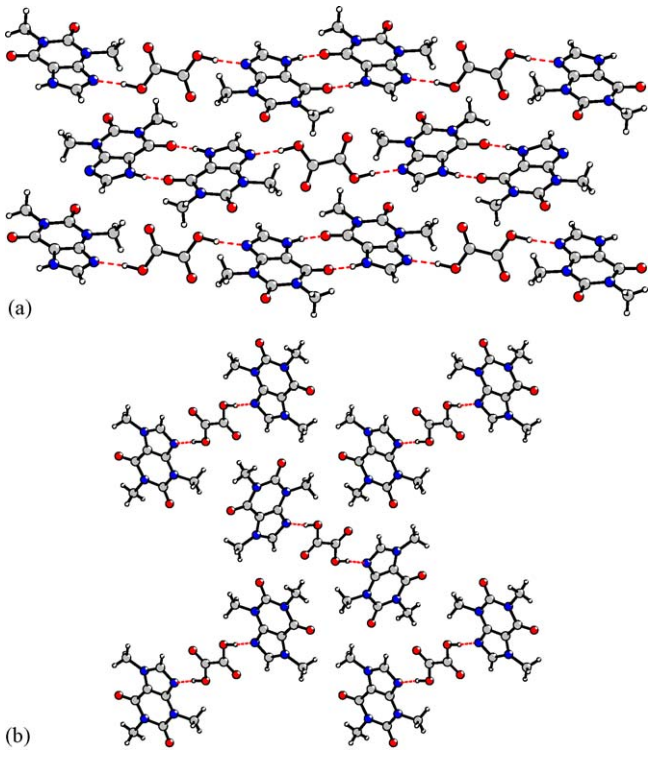


Fig. 2. Crystal packing diagrams of: (a) cocrystal A showing hydrogen-bonded ribbons, and (b) 2:1 caffeine:oxalic acid cocrystal.

However, significant packing differences between the two cocrystal structures appear to arise from the structural variation between caffeine and theophylline. Whereas the caffeine:oxalic acid structure consists of discrete, three-component drug–acid–drug units, the structure of A is comprised of continuous hydrogen-bonded ribbons. These ribbons are enabled by the R_2^2 (10) hydrogen-bonded homo-dimers of theophylline, which are present in the known crystal structure of theophylline monohydrate (Fig. 1b) but are not possible with caffeine cocrystals due to its extra methyl group. This theophylline dimer formation is a persistent feature of each theophylline cocrystal obtained in this study.

A second difference between cocrystal A and the caffeine:oxalic acid cocrystal is that in the caffeine:oxalic acid structure, the caffeine and acid molecules are planar with respect to each other, allowing formation of a weaker intermolecular C–H···O hydrogen bond in addition to the stronger O–H···N hydrogen bond. These two hydrogen bonds together create a caffeine:acid intermolecular hydrogen-bonded ring that may be described in graph set notation as R_2^2 (7). In contrast, the theophylline and oxalic acid molecules in A are not co-planar, and the analogous ring is not formed. This appears to be a result of the presence of the good N–H proton donor on theophylline. Rather than forming the weak C–H···O bond, the oxalic acid carbonyl is directed toward a second theophylline molecule to form what appears to be a long N–H···O interaction (N···O distance 3.204 Å). Lacking a strong donor on the caffeine molecule, this interaction was not possible.

3.1.2. Cocrystal B (1:1 theophylline:malonic acid)

The packing of cocrystal B, involving theophylline and malonic acid, demonstrates the primary intermolecular O–H···N

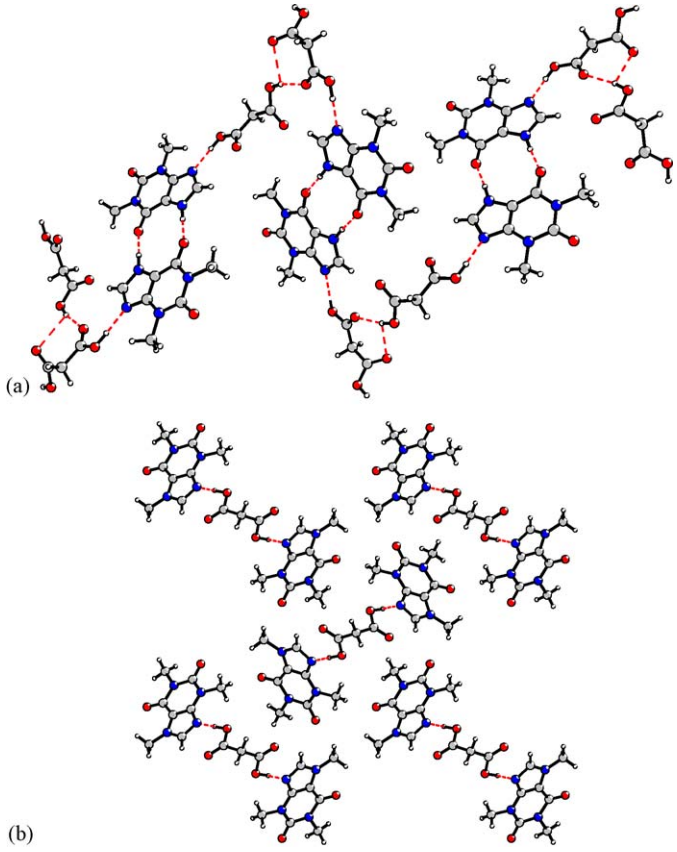


Fig. 3. Crystal packing diagrams of: (a) cocrystal B and (b) 2:1 caffeine:malonic acid cocrystal.

bonds as well as the theophylline dimer formation via secondary N–H···O bonds (Fig. 3a). Aside from those features, the overall packing was surprising in that a 1:1 stoichiometry was observed. This contrasts with the caffeine:malonic acid structure, which exists as a 2:1 cocrystal (Fig. 3b). The 1:1 stoichiometry of B means that only one of the acid groups bonds to a theophylline, leaving the other to participate in a bifurcated hydrogen bond with the two carbonyls of a neighboring acid. The inter-acid hydrogen bond results in a step and a twist away from the plane of each successive theophylline dimer.

3.1.3. Cocrystal C (1:1 theophylline:maleic acid)

Cocrystal C involves the complexation of theophylline with maleic acid. From the crystal structure (Fig. 4a) it is apparent that one maleic acid is present per molecule of theophylline, as was the case with a previously reported caffeine cocrystal (Fig. 4b), though a 2:1 caffeine:maleic acid structure was also reported to form (Trask et al., 2005). In cocrystal C, the R_2^2 (7) hydrogen bond motif between the drug and carboxylic acid is observed, while the second carboxylic group is predisposed to form an intramolecular hydrogen bond as a result of the *cis* geometry of maleic acid. The crystal structure of C also demonstrates the hydrogen-bonded dimerization of theophylline molecules.

3.1.4. Cocrystal D (1:1 theophylline:glutaric acid)

As is true for all theophylline cocrystals described here, cocrystal D demonstrates the expected O–H···N

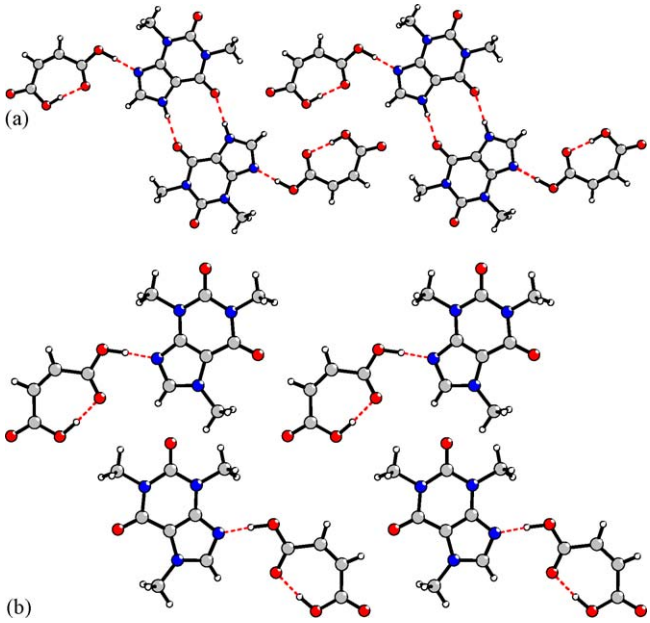


Fig. 4. Crystal packing diagram of: (a) cocrystal C and (b) 1:1 caffeine:maleic acid.

acid–theophylline hydrogen bond as well as hydrogen-bonded theophylline dimerization. While the analogous caffeine:glutaric acid cocrystal was found to be polymorphic (Trask et al., 2004), only one crystal form of the theophylline:glutaric acid system was observed in the course of this work. The packing comparison between D and caffeine:glutaric acid form I (which shares the same primary hydrogen bonding as its polymorph) reveals the similarity of these two analogous cocrystals (Fig. 5). Despite the presence of theophylline dimers, the same drug–acid and acid–acid hydrogen bonds persist in both structures, although the planar sheets of the caffeine cocrystal give way to kinked sheets in D.

3.2. Additional cocrystallization attempts

In addition to the acids depicted in Table 1, cocrystallization of theophylline was attempted with other pharmaceutically acceptable dicarboxylic acids including succinic acid (butanedioic acid), fumaric acid (*trans*-butenedioic acid) and L-tartaric acid (2,3-dihydroxybutanedioic acid). No evidence of cocrystal formation was apparent following attempted cocrystallization via solid-state grinding or solution growth. Similar results were found in a search for two-component caffeine:dicarboxylic acid cocrystals (Trask et al., 2005), suggesting that the cocrystallization preferences of a close molecular analogue of a given API may also provide an indication of which counter-molecules will *not* form cocrystals with that API.

3.3. Relative humidity (RH) stability comparison

A comparison of the RH stability of theophylline cocrystals A–D to that of theophylline anhydrate was performed in order to assess whether these cocrystals offered enhanced physical

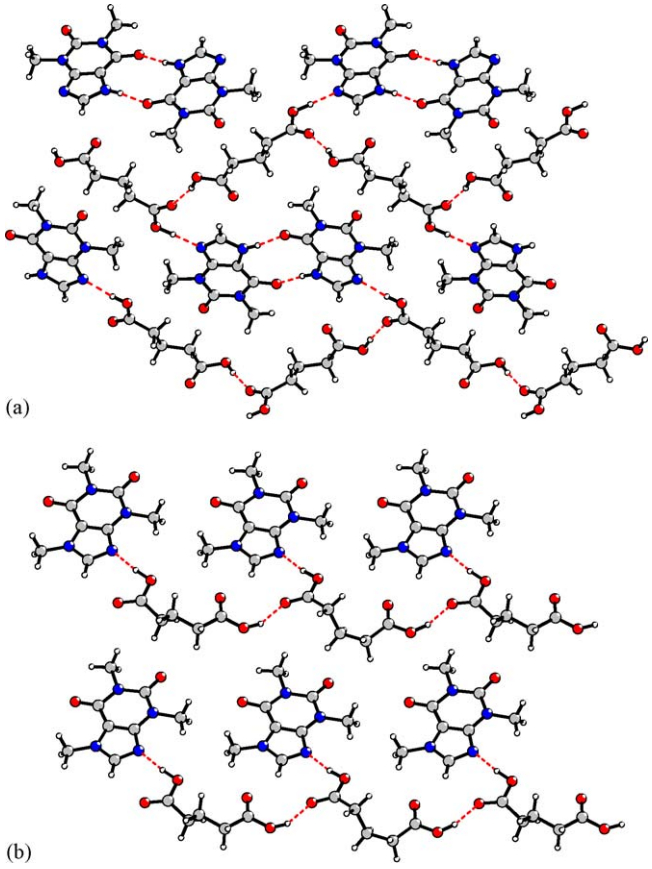


Fig. 5. Crystal packing diagrams of: (a) cocrystal D and (b) 1:1 caffeine:glutaric acid form I.

Table 3
Observed RH stability of theophylline cocrystals

Material	Condition (% RH)	Observed relative humidity stability[a]			
		1 day	3 days	1 week	7 weeks
Theophylline	0	√	√	√	√
	43	√	√	√	√
	75	√	√	√	√
	98	×	×	×	×
Cocrystal A	0	√	√	√	√
	43	√	√	√	√
	75	√	√	√	√
	98	√	√	√	√
Cocrystal B	0	√	√	√	√
	43	√	√	√	√
	75	√	√	√	√
	98	×	×	×	×
Cocrystal C	0	√	√	√	√
	43	√	√	√	√
	75	√	√	√	√
	98	×	×	×	×
Cocrystal D	0	√	√	√	√
	43	√	√	√	√
	75	√	√	√	√
	98	√	×	×	×

[a] *Note*: The symbol (√) indicates that the crystalline material was stable at that condition and time point. The symbol (×) indicates that the crystalline material exhibited physical instability at that time point (see text for details).

stability profiles. The RH challenges, described in detail in Section 2, comprised the storage and subsequent PXRD analysis at four specific RH levels (0%, 43%, 75% and 98% RH) across four different time points (1 day, 3 days, 1 week and 7 weeks). Results are presented in Table 3 and Fig. 6. This stability evaluation procedure mirrors that performed for caffeine anhydrate and its reported cocrystals (Trask et al., 2005), allowing for a direct comparison between the two similar series.

The exact physical stability of theophylline anhydrate and monohydrate with respect to RH is a matter of some disagreement in the literature (Zhu et al., 1996; Ticehurst et al., 2002). Over the course of our 7-week study, we found that at 75% RH and below, theophylline anhydrate was physically stable, while at 98% RH theophylline anhydrate converted into theophylline monohydrate.

In considering the results of the theophylline cocrystals, one observation of significant interest was that in no case was a hydrate of a given theophylline cocrystal observed to form. This may provide evidence that hydrate formation in cocrystals is less likely than in single-component systems; however, one obvious exception to this possible trend is a previously reported monohydrate cocrystal of theophylline and 5-fluoroacetic acid, CSD reference code ZAYLOA (Zaitu et al., 1995). Of additional interest, it was found that cocrystal A was stable up to and including 98% RH. Despite the lack of crystalline hydrate formation in any of the theophylline cocrystals, B, C and D demonstrated similar RH stability to that of theophylline anhydrate, in that they were stable at 75% RH and below. Rather than conversion to a cocrystal hydrate, however, these three cocrystal materials demonstrated dissociation at high RH into theophylline monohydrate and acid (though the acids, with greater apparent aqueous solubility, were less noticeable by PXRD analysis). This dissociation behavior appears to suggest that water successfully competes with malonic, maleic and glutaric acids as a hydrogen bond donor with theophylline, thereby displacing the acidic counter-molecule to preferentially form the crystalline theophylline monohydrate above a critical RH value.

It is noted that an identical overall RH stability profile was observed for each analogous caffeine cocrystal reported recently. The reasons for this similarity of behavior are not yet understood, although it is noted that the strongest acid of each series, oxalic acid (as judged by aqueous pK_a values (O'Neil, 2001)) forms the cocrystals that are most stable with respect to RH. This suggests that in designing cocrystals of an API for enhanced physical stability, the strength of the acid–base interaction may be an important factor. If, with additional study of cocrystal stability behavior, this observation were to develop into a more general trend, this factor might limit the appeal of cocrystal formation involving non-ionic APIs that lack appreciable acidic or basic character. Clearly, further work is needed to evaluate the generality of this trend and to identify other factors that contribute to the increased stability of the theophylline:oxalic acid and caffeine:oxalic acid cocrystals.

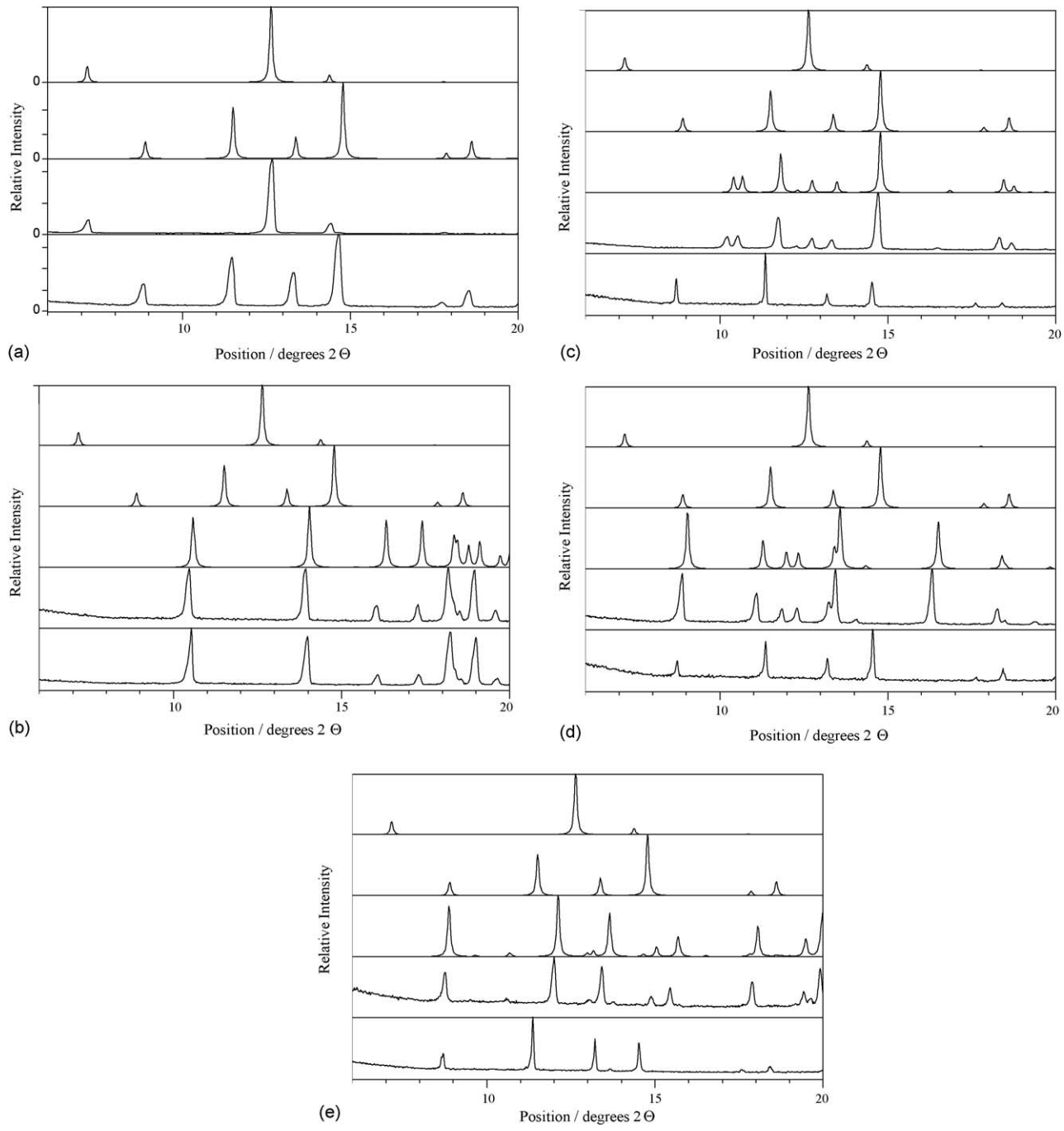


Fig. 6. Overlays of PXRD patterns resulting from the storage of materials at different RH values at the final (7-week) time point of the study. In each overlay, the top two patterns are, from the top: theophylline anhydrate simulated from CSD reference code BAPLOT01; theophylline monohydrate simulated from CSD reference code THEOPH01. The remaining PXRD patterns in each overlay (a–e), from top to bottom, are as follows: (a) result of storing theophylline anhydrate at 75% RH; result of storing theophylline anhydrate at 98% RH; (b) cocrystal A simulated from crystal structure data obtained in this study; result of storing cocrystal A at 75% RH; result of storing cocrystal A at 98% RH; (c) cocrystal B simulated from crystal structure data obtained in this study; result of storing cocrystal B at 75% RH; result of storing cocrystal B at 98% RH; (d) cocrystal C simulated from crystal structure data obtained in this study; result of storing cocrystal C at 75% RH; result of storing cocrystal C at 98% RH; (e) cocrystal D simulated from crystal structure data obtained in this study; result of storing cocrystal D at 75% RH; result of storing cocrystal D at 98% RH.

3.4. Additional investigation of cocrystal A stability

Despite the apparent humidity stability of cocrystal A up to and including 98% RH, when slurried in water (75 mg/ml) for 1 day at ambient temperature, this material exhibited dissociation and formation of theophylline hydrate, as observed by PXRD analysis. This behaviour differed from that of the 2:1 caffeine:oxalic acid cocrystal, which was stable when slurried in water under the same conditions. In considering what might underpin the observed differences between the two analogous cocrystals, two possible hypotheses were envisaged. One explanation considered was that the critical RH necessary to

induce dissociation of cocrystal A was between 98% and 100%, whereas the caffeine:oxalic acid cocrystal exhibited full stability up to and including 100% RH. A second possibility considered was that the mechanism of dissociation of the two cocrystals differed, whereby dissociation of A was mediated by liquid water, and therefore did not occur by exposure only to water vapour.

In order to assess these hypotheses, RH stability evaluation was conducted at 100% RH for both cocrystal A and the caffeine:oxalic acid cocrystal. Upon storing both materials at 100% RH for 1 day, PXRD analysis revealed that while the caffeine:oxalic acid material was unchanged, cocrystal A exhibited peaks corresponding to theophylline hydrate. Furthermore, at 1 week, the cocrystal A material had largely dissociated, while the caffeine:oxalic acid cocrystal remained physically intact. While the mechanisms of dissociation of these two cocrystals are not understood at present, it has been shown that cocrystal A dissociates in the presence of atmospheric humidity, and that the barrier to dissociation lies between 98% and 100% RH. Additional studies, including solubility and hygroscopicity evaluation, are currently underway to further characterize the differences between these two analogous cocrystals.

3.5. Conclusions

This pharmaceutical cocrystallization study involving the API theophylline represents an extension of the reported cocrystallization preferences of a molecular analogue, caffeine. In an industry that often considers molecules in series of small successive chemical modifications, the transfer of "cocrystallization preference" information between analogues may assist pharmaceutical cocrystal design. The observed RH stability behavior of the theophylline cocrystals demonstrates that physical property improvement, and specifically avoidance of hydrate formation, may be achieved via pharmaceutical cocrystallization. The importance of understanding the factors that contribute to the enhanced stability of cocrystal A (theophylline:oxalic acid) are highlighted in light of the previously reported enhanced stability of the caffeine:oxalic acid cocrystal. The observed dissociation behaviour of cocrystal A upon slurry in water represents an important difference between the theophylline and caffeine cocrystal series; this aspect is the subject of ongoing investigation.

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