Switching Polymorph Stabilities with Impurities: A Thermodynamic Route to Benzamide Form III.

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ABSTRACT: We investigate the polymorphic behavior of benzamide, the first compound known to exhibit polymorphism, in the presence of small amounts of nicotinamide in the crystallization environment. A previous study by Emmerling et al.1 showed that the presence of nicotinamide promotes the transformation of the thermodynamic polymorph I of benzamide into its metastable polymorph III via mechanochemistry. We show that this transformation is the result of a thermodynamic switch between these two polymorphic forms driven by the formation of solid solutions with a small amount of nicotinamide. The presence of nicotinamide in the crystallization environment promotes the robust and exclusive crystallization of the elusive form III. These results represent a promising route to the synthesis and utilization of elusive polymorphs of pharmaceutical interest.

Molecular crystals are essential constituents of everyday consumer products such as paints, foods or medicines. The quality, safety and efficacy of such products depend greatly on the properties and structure of the crystals exhibited by their active ingredients. Since most compounds can and will crystallize in various forms (known as polymorphs),2 discovering, understanding and controlling polymorphism is a necessity in product development. In the world of pharmaceuticals, over 50% of compounds exhibit polymorphism.3 Whilst some polymorphs are easy to discover and consistently crystallized, others remain elusive and only appear (or disappear) by chance after decades of research on them.4,5 The quest for new drugs requires, amongst other things, the synthesis, crystallization and characterization of new chemical entities in drug discovery and of the scale up and crystallization process design in drug development. Along the journey towards a final drug product, it is common to observe the appearance of metastable polymorphs first which eventually transform to other more stable polymorphs. This solid form evolution has often been linked to an increase in chemical purity. When chemical purity is low, compounds can be hard to crystallize or do so in unwanted forms. As the synthetic routes are improved and the compounds are subjected to crystallization development, their purity increases which may lead to new crystal forms.6

Whilst crystallization is a very selective process (thus widely used as a purification technique), sometimes the tiniest amount of impurities can have a significant impact on the outcome. This is especially true when the impurity has a close molecular similarity with the compound under development. Impurities are well-known to modify crystal habit7,8 impact the kinetics of crystallization9 and/or inhibit nucleation, crystal growth, and conversion kinetics between polymorphs10 thus promoting or preventing the observation of a particular crystal form. The incorporation of impurities in crystal forms is also known to impact crystal properties such as elasticity or hardness.11

Two notorious examples of drugs changing polymorphic forms in the presence of impurities are aspirin and paracetamol. Despite the fact that aspirin has been crystallized in thousands of tones since the 19th century, its second form of was only discovered in 2005 12 when it was crystallized in the presence of impurities (form II can now be consistently produced in the presence of aspirin anhydride).13–15 Paracetamol, similarly, can only be crystallized as its form II in the presence of various impurities one of which being metacetamol.16 Despite the impact that impurities have on polymorphism and crystallization, there are various molecular mechanisms by which they can act some of which are not well understood. Pioneering early work by the Weizmann group showed how through preferential adsorption on specific crystal faces, impurities are able to modify crystal habits but also inhibit
nucleation and allow for polymorph control. More recent work by Liu et al. has shown how adsorption of metacetamol on form I surfaces in the wrong orientation inhibits its nucleation and growth thus allowing form II to crystallize. In that regard, previous work has mostly focused on using impurities to block the nucleation and growth of stable polymorphs in order to allow for the metastable polymorph to appear. Here we explore a different molecular route of action: rather than blocking the nucleation and growth of the stable form, we explore how other impurities may be able stabilize the metastable polymorph through the formation of solid solutions. Understanding whether or not such routes are possible, and the molecular modes of action would allow us to achieve greater control of polymorphism during crystallization.

![Figure 1. Molecular Structure of benzamide (BZM, left) and nicotinamide (NCM, right)](image)

The polymorphism of benzamide (BZM, Figure 1), the first molecular compound ever reported to be polymorphic, was noted by Wöhler and Liebig in 1832 only to be forgotten for over 170 years. Studies on form I (the stable polymorph) abounded and its structure was first reported in 1959. Because of the elusive nature and metastability of the other polymorphs (II and III) and the impossibility to grow large single crystals of good quality suitable for single X-ray diffraction, the solution of the structure of form II and form III had to wait until the advancement of synchrotron radiation and crystal structure solution from powder methods. More recently, the structure of a metastable and disordered form IV has been proposed. The targeted, robust and exclusive growth of crystals of form III (the form of interest in this contribution) still remains a challenge since this form grows poorly and almost concomitantly with form I.

In a recent study by Emmerling and co-workers, a failed attempt to produce 1:1 cocrystals of BZM with its close relative nicotinamide (NCM, Figure 1) by ball-mill grinding led to the observation of form III BZM. The solid state BZM form I to form III conversion was consistently observed in a range of BZM:NCM concentrations. The authors mentioned in their conclusions that “it was assumed the nicotinamide molecules were included in the crystal system of benzamide, triggering formation of benzamide III”. If, rather than staying at the crystal surfaces blocking growth, impurities are incorporated in lattice positions within crystal structures, they form what it is known as solid solutions. Inspired by these results, we ventured to investigate this historically important compound further in order to shed light on the driving force of the BZM form I to form III conversion.

First, we performed ball-mill liquid assisted grinding (LAG) with ethanol of pure BZM and BZM in the presence of NCM in a range of compositions. LAG of pure BZM affords form I whilst LAG of BZM in the presence of NCM (5-30 mol%) results in BZM form III (ESI S2.3.1), consistent with Emmerling’s work. At concentrations of NCM above 30 mol%, diffraction peaks corresponding to solid NCM start appearing suggesting that the solid solubility of NCM in BZM form III lies around that value. When ethanol is replaced with isopropanol, the LAG results do not change. Interestingly, ball mill neat grinding (NG) of pure BZM resulted in pure BZM form III without the need of any NCM at all (ESI S2.3.2). The observation of form III upon NG of pure BZM may be explained as a crystal size effect. Why small amounts of NCM also induce a BZM form III conversion is less clear since it could potentially be due to a combined size reduction and impurity effect.

In order to explore the possibility of solid-solution formation and its impact on the stability of forms I and III of BZM, we performed lattice energy calculations (PBE-d) for both crystal polymorphs as a function of NCM concentration (Figure 2). For pure BZM, form I is computed to be more stable than form III by only 0.2 kJ/mol. The small energy difference between the forms is consistent with the fact that forms I and III often crystallize concomitantly, with form I known to be the most stable. As the concentration of NCM in the lattice increases, the relative stability of the two polymorphs changes and above 10 mol%, BZM form III becomes more stable than BZM form I. The changes in the lattice energies are, however, very subtle. We note that, since the lattice energies of forms I and III are so close, no computational method available would be able to provide energy differences of such accuracies (within 0.2 kJ/mol). To confirm the revelations of the above simulations, the predicted forms stability changes were assessed experimentally by means of solvent-mediated phase transformation experiments.
If slurry experiments are given enough time, the lower solubility of the more stable form at each given composition would eventually lead the system to thermodynamic equilibrium and thus the more stable form at such composition.

![Figure 2. Lattice Energies of form I and form III of BZM as a function of the NCM concentration.](image)

Excess solid of BZM form I was stirred in saturated isopropanol (IPA) solutions at 25°C in the presence of various amounts of NCM (ESI S2.4.2). 2.5 g of mixed BZM form I with NCM form I solids (at 2, 4, 6, 10 and 20 mol% content of NCM) were added to 5 g of IPA and stirred for one week at which point the solids were characterized. Figure 3 shows the PXRD results for the excess solids of such slurries. Remarkably, the slurry experiments lead to the switch in forms from form I to form III in the presence of NCM. Under these experimental conditions, a total of 4 mol% NCM was required to observe a form I to form III switch in the slurries. When the slurry experiments were repeated in ethanol, the form III conversion was also observed but at slightly higher total concentrations of NCM (7 mol%). These experiments were also performed at higher temperatures, 45°C, with similar results obtained (ESI S2.4.1). The amount of impurity present in the slurry had an important effect on the kinetics of the conversion with kinetics accelerating at higher total concentrations of NCM. For example, slurries from ethanol show that when the concentration of NCM is approximately 10 mol%, the conversion to BZM form III starts after 30 min and completes within 4 h whilst when the NCM concentration is approximately 20 mol% the conversion to form III completes in just 15 minutes (ESI S2.4.1). In all cases, the resulting BZM form III was confirmed to be a solid solution with NCM - as suggested by the absence of diffraction peaks of pure NCM forms, shifts in PXRD peaks and changes in the DSC thermographs. Increasing the concentration of NCM resulted in a linear depression of the melting point of BZM form III from 125.8 °C (10 mol % NCM) to about 121.2 °C (30 mol% NCM) (ESI S2.4.1). Crash cooling experiments (to 25°C, 1.2 supersaturations) of BZM in the presence of NCM (approximately 10, 20 and 30 mol%) were also performed in isopropanol. Consistently with the LAG and the slurry experiments, BZM form III was also obtained via crash cooling in the presence of NCM at all concentrations studied (ESI S2.5).

In the presence of NCM, thus, we are able to consistently and exclusively obtain well-formed form III BZM crystals without the presence of form I crystals, something never achieved before from solution crystallization. This can be achieved both via solvent-mediated phase conversion of form I into form III (slurries) or via crash cooling from solution in the presence of NCM (Figure 4). We notice that the morphologies of our form III are more equant and similar to those of pure form I and very different to the needles of pure form III BZM.24
Figure 3. PXRD patterns of slurry products obtained from mixtures of BZM and NCM in isopropanol at different total concentrations (BZM:NCM in mol% in brackets). Patterns of pure forms are given for comparison (peaks indicated with hashes are typical of BZM-I whilst peaks indicated with asterisk are typical of BZM-III).

Figure 4. Micrographs of form III BZM crystals obtained from slurries (a) and from crash cooling (b) in IPA (20mol% of NCM content).

Until now, we have referred to the amount of NCM content (mol%) with respect to the total amount of solids (BZM+NCM) used in the experiments. In order to estimate the NCM threshold which results in the thermodynamic lattice energy switch (Figure 2), we require to characterize the amount of NCM incorporated in forms I and III BZM crystals. This was quantified by retrieving the solids from the slurry experiments in IPA and characterising them with 1H-NMR. The fraction of the NCM incorporated in the crystals over the total NCM fraction in the experiment (in mol%) was calculated to be 0.7 and 0.8 for BZM forms I and III respectively (see ESI). Thus, NCM incorporates with very equal efficiency in both lattices. We observe that at around 3mol% of NCM incorporation in the BZM crystals, form I starts converting to form III, thus the concentration threshold for the stability switch lies around that value. The discrepancy observed between the calculated 10 mol% and the experimental 3 mol% limits can understandably be attributed to the difficulty of simulating energy changes that are so subtle.

The comparison of the crystal packing of BZM forms I and III reveals a high similarity, with both structures featuring a common 2-D arrangement of centrosymmetric hydrogen-bonded dimers propagating along the a and b crystallographic directions and connected via a second set of N-H…O hydrogen bonds and other non-directional contacts (ESI S2.2). The only difference between structures occurs along the c direction, where the common 2-D arrangements are packed differently and connected via different sets of weaker contacts. The stability switch of the forms in the presence of NCM is driven by two concomitant but opposite effects: i) small destabilization experienced by BZM form I and ii) the increase of stability of BZM form III upon incorporation of small amounts of NCM. Given the high structural similarity between the two polymorphs, the incorporation of NCM in the crystal lattice is only subtly impacting weak and non-directional contacts. In this case, the “switch” in stability takes at very small impurity incorporations (3 mol%) because of the close relative lattice energy of the two pure polymorphs.

In conclusion, we have demonstrated that the relative stability of polymorphs can be inverted by using selective impurities able to form solid solutions. This has been shown for benzamide forms I and III in the presence of nicotinamide. Lattice energy calculations performed on both crystal polymorphs as a function of NCM content...
suggested a change in the relative stability of the two polymorphs as the concentration increases. This observation was confirmed experimentally from results of solvent mediated phase transformations that show full conversion from BNZ form I to BNZ form III starting from concentrations of NCM in the solid lattices of just 3 mol%. The concentration of NCM in the solution was shown to impact the kinetics of the form I to form III conversion, increasing the transformation rates at increasing concentrations of NCM. PXRD, NMR and thermal analysis confirmed the formation of solid solutions. Although impurities are well-known to impact crystal habit, slow nucleation and crystallization kinetics and impact polymorphism, their role in forming solid solutions and the resulting impact on polymorphism has been explored only rarely. To the best of our knowledge, this study shows for the first time experimentally and computationally that impurities can invert the thermodynamic stability of polymorphs through the formation of solid solutions. This mechanism may be able to explain the appearance and disappearance of some polymorphs. Through its understanding, we are now seeking to rationally design such impurities in order to achieve reliable access of elusive or computationally predicted polymorphs though this type of “solid-solution thermodynamic switch”. This novel concept opens a new route to new polymorphs and their control, which will be of major interest to the pharmaceutical industry.

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