Journal Name

ARTICLE

Isomorphous Template Induced Crystallisation: A Robust Method for the Targeted Crystallisation of Computationally Predicted Metastable Polymorphs.

Vijay K. Srirambhatla,[a] Rui Guo,[b] Sarah L. Price[b] and Alastair J. Florence*

A new method of inducing the crystallisation of metastable polymorphs by isomorphous templating has been developed and used to reproduce the crystallisation of CBZ-V on the surface of DHC-II. Studies of the growth of CBZ-V on DHC-II single crystals show crystals growing laterally and vertically on DHC-II surfaces without any significant face selectivity. The generality of this computationally inspired crystallisation approach is demonstrated by producing the first crystals of an entirely new polymorph of cyheptamide, which is isomorphous to both DHC-II and CBZ-V.

Polymorphism[1] is the ability of a compound to adopt different crystal packing arrangements. Whilst this can pose a challenge for industrial crystallisation processes to deliver consistency, polymorphs also present the opportunity to vary structure–property relationships including crystal shape, solubility, stability or mechanical properties. Whilst solution based polymorph screening[2] is routinely performed to identify all accessible forms, these tend to rely on trial and error variation of crystallisation conditions to find appropriate nucleation and growth conditions that promote alternative forms. Whilst surface heteronucleation[3] or templating has been demonstrated using polymers[4], crystals[5] and other materials, the mechanisms by which a specific form is obtained are poorly understood. Crystal structure prediction methods and targeted experimental approaches can offer a possible route to target the first crystallisation of specific thermodynamically-plausible polymorphs.
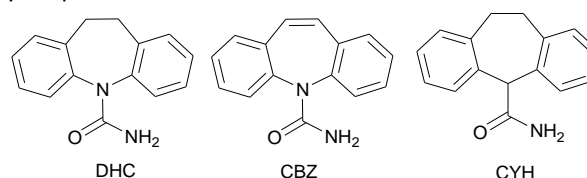

Figure 1: Molecular structure of carbamazepine (CBZ), dihydrocarbamazepine (DHC) and cyheptamide (CYH).

An experimental approach informed by crystal structure prediction (CSP) methods[6] led to a new predicted, and previously unobserved, metastable polymorph of carbamazepine (CBZ; CBZ-V).[7] CBZ-V crystals were obtained from vapour phase deposition of CBZ onto dihydrocarbamazepine (DHC) form-II crystals, isomorphous to the predicted form of CBZ-V. However, this raised several important questions: 1. Was the appearance[8] of CBZ-V crystals on DHC-II crystals a case of a disappearing polymorph? Initial attempts to reproduce form V on isolated DHC-II single crystals were not reliable. 2. Is there any alternative facile method to template the formation of CBZ-V crystals? 3. Is there any relationship between the orientation of the DHC-II and CBZ-V crystals? 4. Since DHC-II and CBZ-V are isomorphous, epitaxial growth of CBZ V on DHC II would be expected to give rise to a uniform layer of growth of CBZ-V around the DHC-II seed. Is this process observed? 5. Finally, and crucially, can the computationally assisted method be applied to discover predicted thermodynamically feasible polymorphs of other compounds that are not found in routine screening?

Here we report a robust template-induced crystallisation method for the crystallisation of CBZ-V and demonstrate the versatility of the method by targeted crystallisation of a new predicted form of cyheptamide (CYH). To find CBZ-V more reliably, a polycrystalline template method has been developed.

a. Strathclyde Institute of Pharmacy and Biomedical Sciences, University of Strathclyde, Technology Innovation Centre, 99 George Street, Glasgow, G1 1RD, UK. E-mail: alastair.florence@strath.ac.uk; Fax: 0141-552-2562; Tel: 0141-548-4877.
b. Department of Chemistry, University College London, 20 Gordon St, London, WC1H 0AJ, UK.

A thin polycrystalline layer of DHC-II was obtained by drop casting a slurry of DHC in ethylacetate onto a glass slide and allowing the residual solvent to evaporate at room temperature (ESI). This thin polycrystalline layer of DHC-II was used as a template surface for vapour deposition experiments. Around 40 mg of CBZ was placed in a petri dish which was placed on a heating plate maintained at around 125 °C. The polycrystalline template slide was placed ~1 cm above the CBZ such that CBZ vapour would come into direct contact with the DHC-II layer (ESI). The deposition of CBZ on the polycrystalline DHC-II template was allowed to continue for about 48 hours. Clear evidence of crystal growth was observed and surface grown crystals were analysed by X-ray powder diffraction, confirming the presence of both templated CBZ-V and non-templated CBZ-I crystals (ESI). Individual needle shaped crystals were confirmed as CBZ-V using single crystal X-ray diffraction.
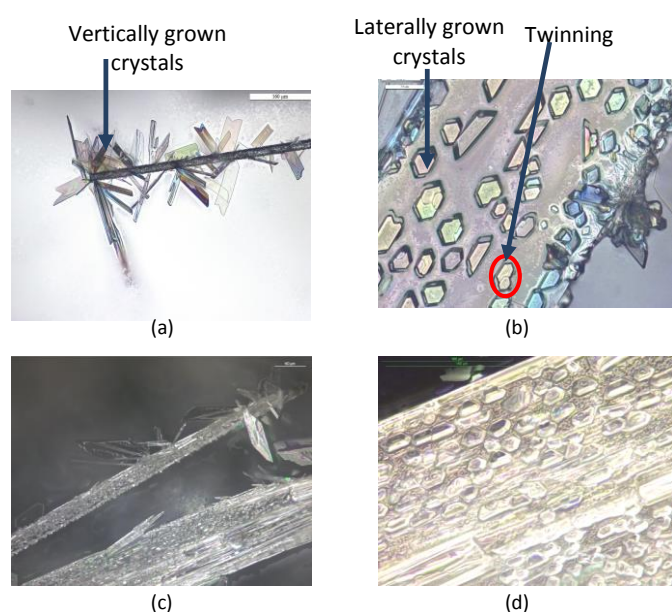

(a) (b)

(c) (d)

Figure 2: Optical microscope images of CBZ-V on DHC-II showing (a) vertically (b) laterally grown CBZ-V crystals; (c) & (d) vertically and laterally grown CYH-III crystals on DHC-II surface, respectively.

The polycrystalline DHC-II template significantly increases the surface area and range of orientations of different crystal facets presented for vapour deposition (ESI). To investigate the templating effect between the substrate and deposited crystals further, a new template was prepared comprising an ordered array of individual single crystals of DHC-II mounted on a glass slide. The vapour deposition experiments were repeated and CBZ crystals were found to grow both laterally and vertically on the DHC-II template crystal surfaces (Figure 2). No apparent face selectivity of any of the deposited crystals was observed from any of the experiments. Vertically oriented CBZ crystals are observed growing in clusters from template surface features similar to those previously reported[5]. Whilst the growth of

some of these vertical crystals could be due to defects or ledges on the DHC-II surface, in some instances it was also observed that the laterally grown microcrystals also promote the growth of vertical crystals. Although only lateral growth of microcrystals was observed, it would be expected that with sufficient time, individual microcrystal boundaries would merge creating a complete surface layer of CBZ-V on the DHC II seed. It is also interesting to note evidence of growth twinning amongst lateral grown crystals (Figure 2(b)). The presence of two distinct populations of deposited crystals (small lateral and large vertical) suggests two crystallisation mechanisms are taking place during the deposition experiments. Specifically, these results are consistent with epitaxial growth of CBZ-V on DHC-II leading to the lateral microcrystals via a birth and spread mechanism[9], plus the templated heterogeneous nucleation of vertically grown CBZ-V from DHC-II surface ledges.
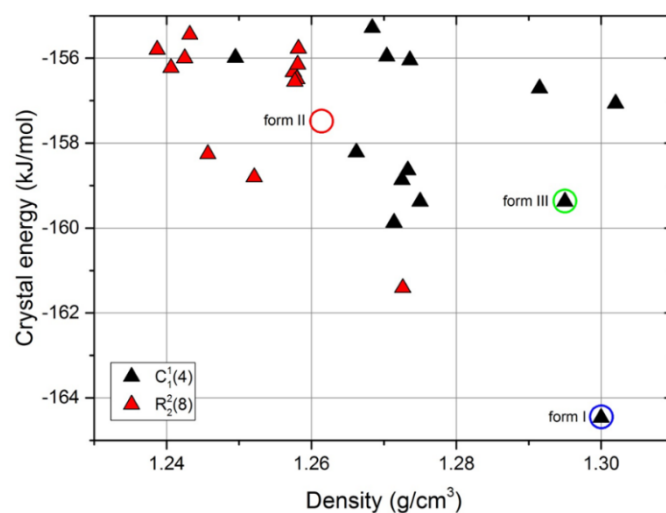

Figure 3: Crystal structure prediction (CSP) study of CYH, where each triangle corresponds to a CSP generated mechanically stable crystal structure, coloured by whether it contains a hydrogen bonded dimer or catemer. The crystal energies are the estimated Helmholtz free energies at 298 K, calculated in a polarisable continuum. The circles denote the structures corresponding to the three experimental CYH polymorphs, including CYH-III from this work.

To further test the utility of the new experimental templating method a crystal structure prediction (CSP) study was performed for CYH (Figure 3). The CSP study used Crystal Predictor[10] to generate Z'=1 crystal structures in 60 space groups, allowing for full rotation of the amide torsion angle. The structures were then refined using CrystalOptimizer[11] to allow all the flexible torsion angles to adapt to the packing forces. This was done by optimising the lattice energy, calculated as a sum of the intramolecular energy penalty, calculated from the isolated molecule PBE0/6-31G(d,p) charge density, and the intermolecular energy calculated from the corresponding distributed multipoles[12] and the empirical *exp-6* FIT potential[13]. The effect of recalculating the charge density in a polarizable

continuum with ε=3, typical of organic crystals[14], and of including a rigid-body harmonic estimate of the thermal effects[15,16] was investigated. The relative energies of the three CYH polymorphs were also calculated by periodic density functional calculations using two dispersion corrections[17,18] in CASTEP[19]. Further details are in the ESI. The CSP study indicated that a polymorph of CYH, isostructural to CBZ-V and DHC-II is thermodynamically feasible and potentially more stable than CYH-II (ESI).

Table 1: Unit cell variations of CYH-III (predicted and experimental), CBZ-V and DHC-II.

Pbca	a/Å	b/Å	c/Å	α, β, γ/°
CYH CSP#16	9.2878	11.0190	23.8845	90
Experimental CYH form III	9.0171(5)	11.0071(6)	23.9483(13)	90
DHC form II (VACTAU02)	9.0592(4)	10.3156(5)	25.0534(12)	90
CBZ form V (CBMZPN16)	9.1245(5)	10.4518(5)	24.8224(11)	90

The polycrystalline DHC-II template approach was applied to CYH and successfully obtained a new polymorph, CYH-III. The single-crystal template approach was used to produce samples suitable for structure determination (Figure 2; ESI). The crystal structure confirmed the new form was isomorphous with the computationally predicted polymorphic form of CYH (Table 1 (CSP#16;); Figure 4)‡. As in CBZ-V and DHC-II, the amide groups in CYH-III form hydrogen bonded catemers through N-H···O interactions that extend along the [010] direction. In addition, the CYH-III crystals adopt the same lateral and vertical orientations on DHC-II templates suggesting the same crystallisation mechanisms to those producing CBZ-V are involved.
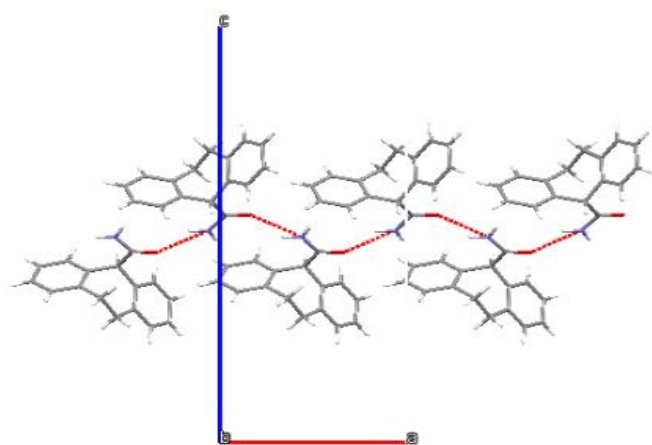

Figure 4: Crystal packing of CYH-III viewed down *b*.

The molecular structures of DHC, CBZ and CYH (Figure 1) show that CYH differs more in molecular shape from DHC than CBZ

due to the geometry around the sp^3 C in the central 7-membered ring. This difference in molecular structure may explain why it has not been possible to form a 1-1 solid solution of CYH:DHC, whereas a solid solution of CBZ:DHC can be readily obtained via slow evaporation from ethylacetate solution.[20] Despite this difference in molecular structures, the targeted growth of CYH-III on DHC-II occurs highlighting the flexibility of the computationally assisted template induced crystallisation method.

Whilst the new experimental method has proven to be more robust than using isolated single crystals as templates, understanding the mechanisms of isomorphous templating remains challenging. We observe a variety of different growth modes and morphologies, including the nucleation of vertically grown crystals (ESI). The growth of lateral CBZ-V and CYH-III on single crystals of DHC-II and on the polycrystalline material is consistent with an epitaxial mechanism however, uniform layered growth is yet to be observed. The occurrence of vertically aligned crystals also suggest that there is more than one mechanism involved.

In conclusion, by targeted crystallisation using an isomorphous template of DHC-II, we have produced a new polymorph of CYH which was not found in conventional screening methods.[21] CYH-III is computed to be more stable than CYH-II (ESI) by a variety of methods. The adapted experimental templating method using polycrystalline material and multiple single crystals has been successfully applied to CBZ-V and for the discovery of CYH-III. The combination of mechanisms involved suggest that multiple approaches may be developed to mimic and control these and further apply this approach to the discovery of other polymorphs for a wider range of compounds.

Acknowledgements

Notes and references

‡ Diffraction data were collected at 123 K for a CYH-III crystal measuring 0.05 x 0.30 x 0.40 mm using Cu-Kα radiation (λ = 1.54180 Å), measured reflections = 13538, independent reflections = 2414, θ range/° = 6-75, R_{merge} = 0.06, 169 parameters, R = 4.4, (based on F and 1991 data with F2> 3σ (F2)), R_w = 4.6, S = 0.891. Orthorhombic, space group Pbca, unit cell parameters a = 9.0171(5) Å, b = 11.0071(5) Å, c = 23.9483(13) Å, volume = 2376.9(2) (Å)^3, Z = 8, ρ_c = 1.326 g cm^{-3}, $C_{16}H_{15}N_2O_1$, Mr = 237.3.

1. J. Bernstein, *Polymorphism in Molecular Crystals*, Clarendon Press, Oxford, U.K., 2002.

2 A. J. Florence, *Polymorphism in Pharmaceutical Solids*, CRC press, 2nd edn., 2009, ch-5, pp.139-184.

3. R. Davey, J. Garside, *From Molecules to Crystallizers. An Introduction to Crystallisation*, Oxford University Press, Oxford, 2001, pp. 15-25.

4. P. Y. Laura, P. C. Price, J. F. Jessica, A. J. Matzger. *J. Am. Chem. Soc.*, 2015, 137, 871-875.

5 . C. A. Mitchell, L. Yu, M. D. Ward, *J. Am. Chem. Soc.*, 2001, 123, 10830-10839.

6. (a) S. L. Price, *Chem. Soc. Rev.*, 2014, 43, 2098-2111 (b) S. L. Price, D. E. Braun, S. M. Reutzel-Edens Chem. Commun. DOI: 10.1039/C6CC00721J (c) S. L. Price, *Acc. Chem. Res.*, 2009, 42, 117−126.

7. J. B. Arlin, L. S. Price, S. L. Price, A. J. Florence, *Chem. Commun*, 2011, 47, 7074–7076

8. J. D. Dunitz, J. Bernstein, *Acc. Chem. Res.*, 1995, 28, 193-200.

9. M. Ohara, R. C. Reid, *Modeling Crystal Growth Rates from Solution,* Prentice-Hall International Series in the Physical and Chemical Engineering Sciences, 1973, Prentice-Hall, p. 272.

10. M. Habgood, I. J. Sugdan, A. V. Kazantsev, C. S. Adjiman, C. Pantelides, *Journal of Chemical Theory and Computation*, 2015, 11(4), pp. 1957-1969.

11. A. V. Kazantsev, P. G. Karamertzanis, C. S. Adjiman, C. Pantelides, *Journal of Chemical Theory and Computation*, 2011, 7(6), pp. 1998-2016.

12. A. J. Stone, *Journal of Chemical Theory and Computation,* 2005, 1(6), pp. 1128-1132.

13. S. L. Price, M. Leslie, G. W. A. Welch, M. Habgood, L. S. Price, P. G. Karamertzanis, G. M. Day, *Physical Chemistry Chemical Physics,* 2010, 12(30), pp. 8478-8490.

14. T. G. Cooper, K. E. Hejczyk, W. Jones, G. M. Day, *Journal of Chemical Theory and Computation,* 2008, 4(10), pp. 1795-1805.

15. A. T. Anghel, G, M. Day, S. L. Price, *CrystEngComm,* 2002, 4(62), pp. 348-355.

16. G. M. Day, S. L. Price, M. Leslie, *Journal of Physical Chemistry B*, 2003, 107(39), pp. 10919-10933.

17. S. Grimme, *Journal of Computational Chemistry,* 2006, 27(15), pp. 1787-1799.

18. A. Tkatchenko, M. Scheffler, *Physical Review Letters*, 2009, 102(7), p. 073005.

19. S. J. Clark, M. D. Segall, C. J. Pickard, P. J. Hasnip, M. J. Probert, K. Refson, M. C. Payne, *Zeitschrift fur Kristallographie*, 2005, 220(5-6), pp. 567-570

20. A. J. Florence, C. K. Leech, N. Shankland, K. Shankland, A. Johnston, *CrystEngComm,* 2006, 8, 746-747.

21. A. J. Florence, K. Shankland, T. Gelbrich, M. B. Hursthouse, N. Shankland, A. Johnston, P. Fernandes and C. K. Leech, CrystEngComm, 2008, 10, 26-28.