

Disappearing Polymorphs Reappear in the Mill: The Case of Ritonavir

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Summary

Organic compounds can crystallise in different forms known as polymorphs. Some polymorphs have disappeared from the physical world, never to be recrystallised again under the same conditions. The most infamous of these cases is that of the HIV drug ritonavir: once its reluctant stable form II was unwillingly nucleated for the first time, its desired but metastable form I could never be produced again with the same manufacturing process. The disappearance of metastable polymorphs remains a mysterious phenomenon, and the lack of control over it can be frustrating and costly for drug development. Here we show that Ritonavir's extraordinary disappearing polymorph can be consistently produced by ball-mill grinding. Our work shows that not only crystal size, but also crystal shape and molecular conformation dictate polymorph stability switches in the mill. Through population balance modelling simulations, we also demonstrate how the size and shape of crystals at the steady state are determined by crystal breakage, dissolution, and growth kinetics in the mill, which in turn can be controlled by the milling conditions. This work highlights the huge potential of mechanochemistry in polymorph discovery, and the manufacturing and control of polymorphs of complex flexible drug compounds such as Ritonavir.

Tablets are the most common vehicles for drug delivery. They contain powders of active pharmaceutical ingredients (APIs) together with excipients. Although pharmaceutical solids are generally stable and easy to handle, they also face multiple challenges in their journey through pharmaceutical development. Perhaps one of the most important challenge of them all is that of crystal polymorphism,¹ especially when it is late appearing,² disappearing^{3,4} or uncontrolled.⁵ Polymorphism is the ability of a compound to crystallise in more than one crystal structure¹ often also involving significant changes in molecular conformation (conformational polymorphs).⁶ With the ever increasing molecular complexity of modern APIs, controlling the impact of molecular flexibility in polymorphism⁶ and crystallisation^{7,8} is critical to enable the development of modern drugs.

The most infamous example of conformational and disappearing polymorphism is undoubtedly that of ritonavir (RVR).² RVR, an HIV-protease inhibitor antiviral for treating acquired immune deficiency syndrome, was developed and commercialised by Abbot Laboratories in the form of gel capsules formulated based on the knowledge of RVR form I (RVR-I). Two years after market launch, the manufacturing process designed to produce RVR-I yielded the more stable form II (RVR-II), compromising the product's solubility and bioavailability.² Intriguingly, once RVR-II crystal seeds had formed at any of Abbot's laboratories or manufacturing plants, RVR-I could not be obtained any longer. The capsules had to be withdrawn from the market and reformulated after significant efforts from Abbot's scientists.⁹ The RVR molecule is large and flexible with the configuration around several of its rotatable bonds changing in its two polymorphs. For simplicity and because of its significance, we focus our attention on the carbamate group, which adopts a stable *trans* configuration in RVR-I and a metastable (+30 kJ mol⁻¹) *cis* configuration in RVR-II (Figure 1a). The *cis* configuration is not only disfavoured thermodynamically but also kinetically because of the very high energy barrier for the *trans* to *cis* interconversion (~90 kJ mol⁻¹).¹⁰ The difficulties of nucleating RVR-II for the first time have partly been ascribed to a low abundance of the *cis* conformer in solution,² but what would happen if we were not to use solution crystallisation?

In this work we study the polymorphism of RVR by means of neat (NG) and liquid-assisted (LAG) ball-mill grinding.¹¹ First, we explore whether the conversion between RVR-I and RVR-II forms, with

the required conformational changes, is possible by milling. It has been previously shown that the environmental conditions of ball-milling experiments (i.e. the amount of solvent or its nature)¹¹⁻¹⁴ are instrumental for enabling polymorphic transformations in the mill.¹⁵ After some experimental exploration, conditions were found for which RVR-I and RVR-II could consistently interconvert in the mill with conversion kinetics shown in Figure 1b for LAG experiments of RVR with isopropanol (IPA) and water. From RVR-I, a full conversion to RVR-II was achieved after just a few minutes by milling with IPA at $0.25 \mu\text{L}\cdot\text{mg}^{-1}$. This observation is remarkable, considering the difficulty of obtaining crystals of RVR-II from solution for the first time.² From RVR-II, LAG with water enabled conversion to RVR-I but it required of significantly longer milling times (>120 min).

Once conversion kinetics were established, milling equilibrium curves were produced for five solvents (Figure 1c). In these curves, the phase composition of the LAG product at steady state is shown as a function of solvent concentration. Milling with no solvent (neat-grinding, NG, or at low solvent concentrations) always afforded RVR-I. In all tested solvents except water, as the solvent concentration was increased, RVR-II started appearing until it became the sole phase at concentrations above ~ 0.15 - $0.25 \mu\text{L}\cdot\text{mg}^{-1}$. The shallow slopes of the equilibrium curves suggest the presence of an additional third phase in the milling products, in accordance with previous observations^{13,16} and with the theory of cooperativity of Hunter and Anderson.¹⁷ As new diffraction peaks in the PXRD patterns of the products were absent, the third phase must be constituted by amorphous RVR (RVR-A).

Amorphous phase quantification of the LAG product with IPA ($0.10 \mu\text{L}\cdot\text{mg}^{-1}$) was performed via Rietveld analysis with the internal standard method. Under those conditions, the milling powders were composed of $\sim 45\%$ RVR-I, $\sim 30\%$ RVR-II and $\sim 25\%$ of RVR-A (Figure 1d). An SEM image of the milling product showed regions with aggregated particles and regions with dispersed needles. Full amorphisation of RVR was achieved upon NG for over 120 min. RVR-A was produced from both RVR-I and RVR-II and spectroscopic methods confirmed that both RVR-A phases obtained were identical. The FTIR spectra of RVR-I, RVR-II and RVR-A revealed a greater structural similarity of RVR-A with RVR-I suggesting that the more stable *trans* carbamate conformation is also dominant in RVR-A (see SI). The crystallite sizes (Scherrer) of the LAG products in various solvents, calculated for the minimum solvent concentration affording at least 85% of each phase, were found to be around 70 nm and 80 nm for RVR-I and RVR-II, respectively (Figure 1c). Smaller crystallite sizes were found for milling conditions that lead to RVR-I (lower solvent concentration). We note that crystallite sizes for the products in ethyl acetate are particularly small (~ 60 nm) for both polymorphs, suggesting that crystal surfaces might be particularly stabilised in this solvent.

Since polymorphs typically differ in lattice energies by just a few kJ mol^{-1} ,¹⁸⁻²⁰ differences in their surface structures and stabilities can lead to thermodynamic stability switches at the nanoscale.^{13,20} This has been shown to drive polymorphic conversions in the mill.¹⁸⁻²⁰ The energy differences for conformational polymorphs, however, are more significant than packing polymorphs,¹⁸ and we found surprising to observe similar conversions for RVR.

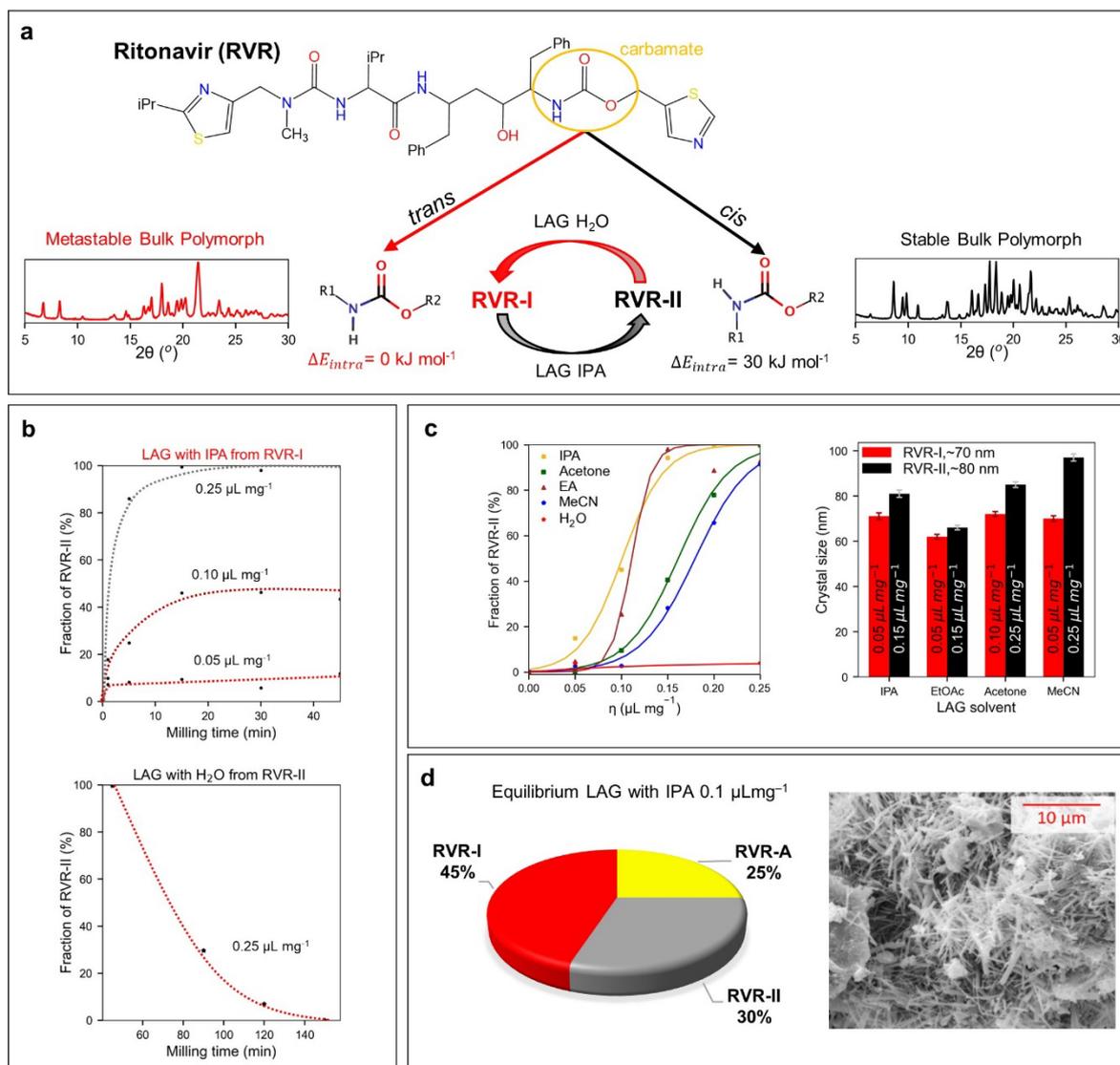


Figure 1. **a**, RVR molecular structure, carbamate configurations and RVR-I and RVR-II interconversion upon milling. **b**, Kinetics of the RVR-I to RVR-II conversion (LAG with IPA, upper) and the RVR-II to RVR-I conversion (LAG with water, lower). **c**, Equilibrium milling curves as a function of solvent concentration (η) in five solvents (left, EA=ethylacetate and MeCN=acetonitrile) and crystal sizes from PXRD data analysis around for RVR-I and RVR-II around the critical solvent concentration. **d**, Phase composition of the equilibrium product of RVR LAG with IPA at 0.10 $\mu\text{L mg}^{-1}$ (left) with SEM image (right). The lines in the plots serve as a guide for the eye. In the plot of panel c, lines were obtained by fitting a sigmoid model.

To explore the thermodynamics of the RVR polymorphs at the nanoscale, we performed state of the art DFT-d calculations of the particle energies of RVR-I and RVR-II as a function of size and shape using equation (1).¹³

$$E_{particle}(r, \phi) = (E_{latt-inter} + \Delta E_{intra}) + (-0.5 \sum_{(hkl)} x_{(hkl)}(r, \phi) E_{att}^{(hkl)}) \quad (1)$$

The first term in the equation is the lattice energy (with an inter and an intra molecular part) and the second term is the surface energy penalty of the particle which can be computed by adding up the individual surface energies of the (hkl) facets weighted by their morphological importance, $x_{(hkl)}(r, \phi)$. The morphological importance depends on the size, r , and shape, ϕ , of the crystal.

Lattice energies for RVR-I and II were computed to be -395 and -400 kJ mol^{-1} respectively, with RVR-II computed to be the stable bulk polymorph. The surface energy penalty of RVR-I and RVR-II was then evaluated in order to compute the particle energy. Figure 2a shows how the energy penalty in RVR-II surfaces is typically higher than the energy penalty of in RVR-I surfaces. To illustrate the impact of the crystal shape on the crystal particle energy, the surface energy penalty for both RVR polymorphs was computed for 300 different crystal habits with L/T and W/T aspect ratios between 1 and 14 (where L, W and T are the length, width and thickness of the crystals) keeping the size of the particle constant at PED 60 nm (Figure 2b, where PED is the size of the diameter of an equivalent sphere with identical volume). We observed a strong dependence of the surface energy penalty with crystal shape. For both forms, the surface energy penalty is at its minimum for blocky crystals, and it increases as the particles become more elongated in either the L or W dimension. In figure 2c, the dependence of the particle energy as a function of size is then shown for both polymorphs adopting two sets of crystal morphologies. The particle energies of RVR-I and RVR-II do not cross when their morphologies are block-like (Figure 2b, left) but they cross for needles (Figure 2b, right). This shows that needles of RVR-I are more stable than needles of RVR-II at small sizes. Finally, we computed the probability of two random crystal morphologies within our defined aspect ratios to cross in energy as a function of PED size ($>100k$ computations). This probability is close to zero at sizes of 100 nm and increases up to 65% as the size decreases to 20 nm. The stability switch is most likely at small particle sizes ($\sim 40\text{nm}$) and for needle morphologies. We note that the latter represent the experimental morphologies typically observed for RVR-I and RVR-II both from solution crystallisation^{9,21} and from our ball-milling experiments (Figure 1d).

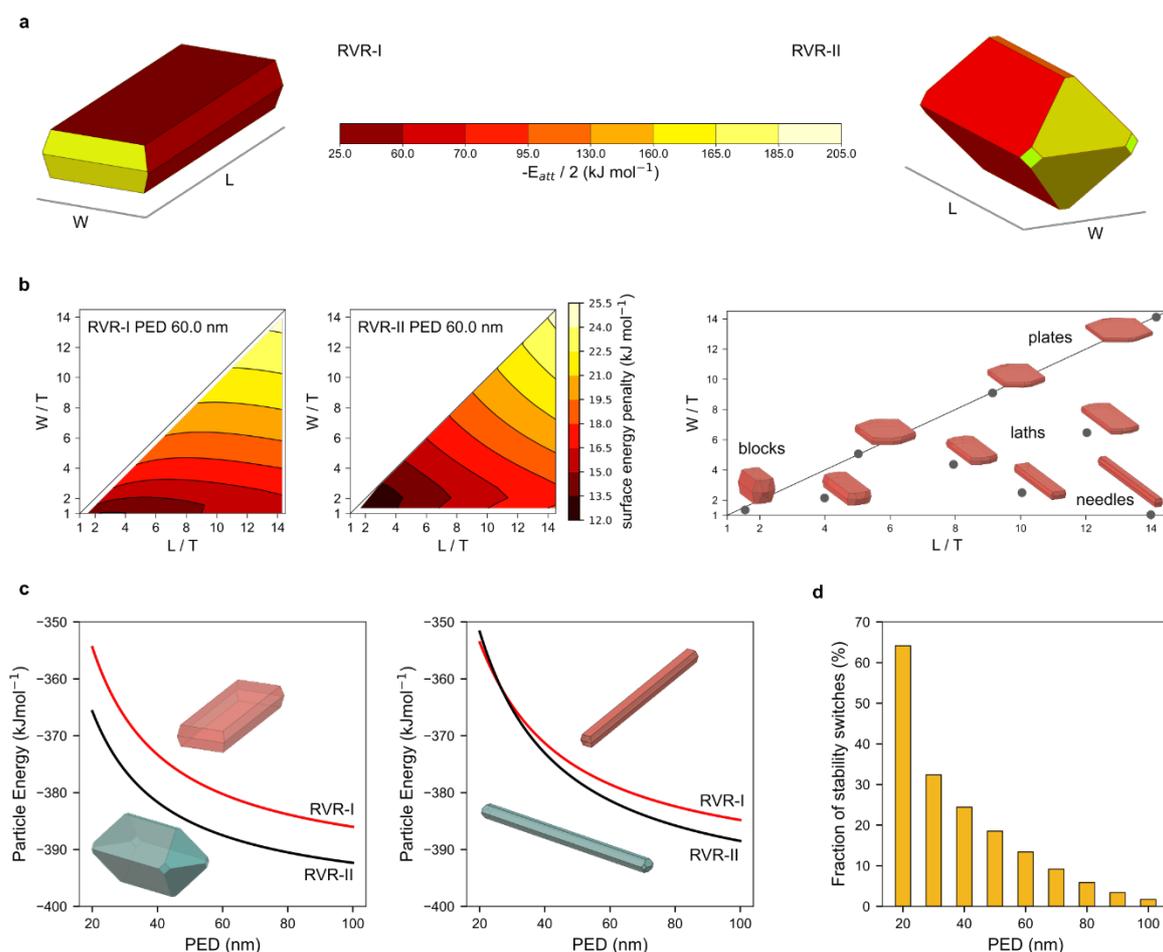


Figure 2. The thermodynamic stability of RVR polymorphs at the nanoscale depends on the relative destabilisation due to crystal surfaces, which in turn is a function of the crystal size and shape. In general, RVR-II has higher surface energies. **a**, Predicted attachment energy morphologies for RVR-I

and II with facets coloured according to their absolute surface energy penalty. **b**, Surface energy penalties for RVR-I and II computed at a constant Particle Equivalent Diameter (PED) of 60 nm as a function of crystal shape (left) and map of corresponding morphologies (right). **c**, Particle energy as a function of size for attachment energy morphologies (left) and for arbitrary needle morphologies (right). **d**, Fraction of combinations of morphology pairs which afford a switch of thermodynamic stability between RVR-I and RVR-II (RVR-I becomes the stable form) at the given particle size.

To aid understanding the processes occurring in the mill, we modelled particle size evolution using population balance equation models (PBEMs) for three potential scenarios (a, b and c in Figure 3).²²⁻²⁴ The column **Concept** in Figure 3 shows the qualitative evolution of the particle size distribution whilst the column **Population Balance Equation Model** shows the corresponding evolution of mean particle size, supersaturation, particle number, and particle volume for the three scenarios.

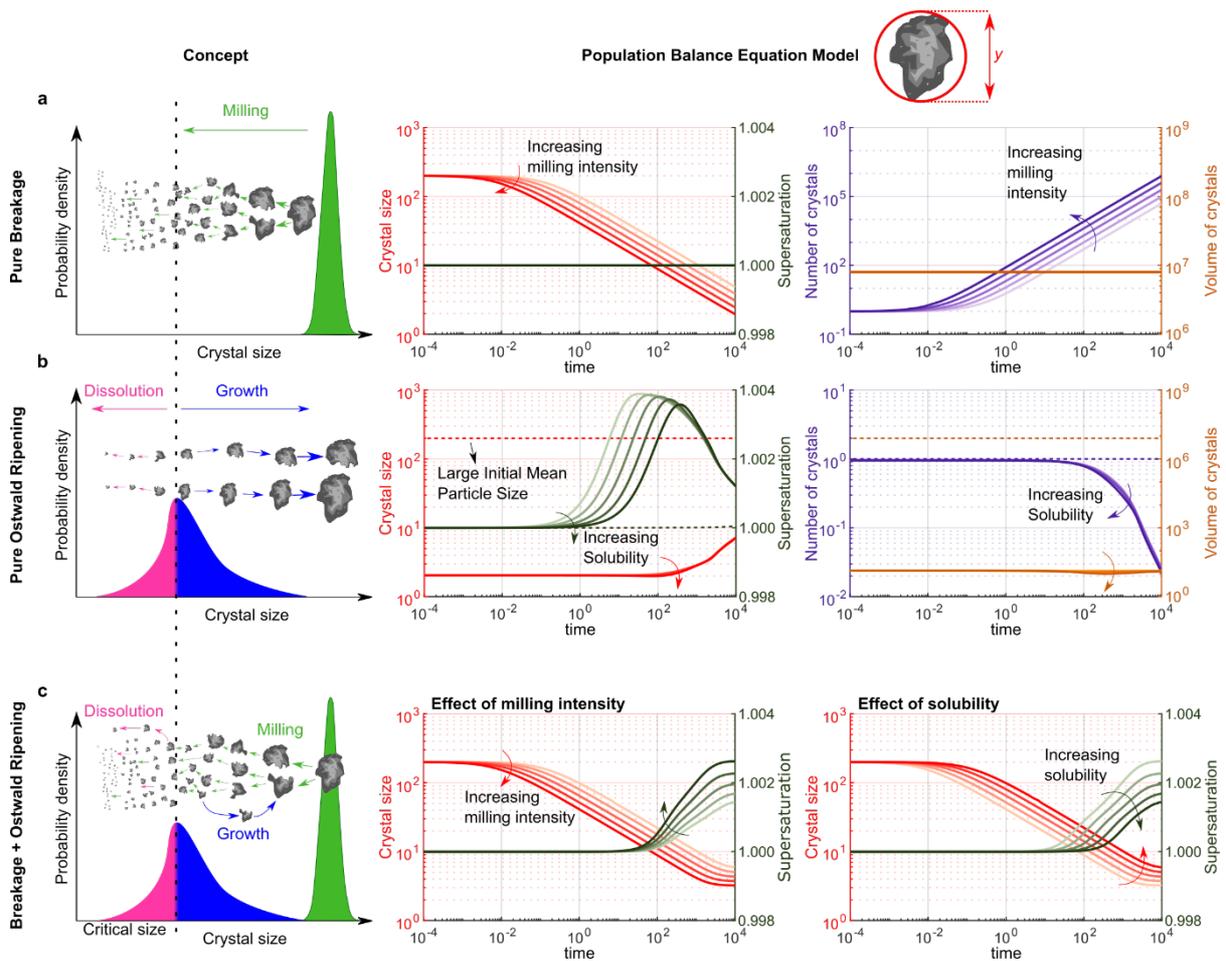


Figure 3. Qualitative evolution of the particle size distribution (column concept) with its corresponding evolution of mean particle size, supersaturation, particle number, and particle volume for the three scenarios studied with PBEM. Scenario a corresponds to pure breakage, scenario b corresponds to pure Ostwald ripening and scenario c to a combination of both processes. The dashed line in column **Concept** indicates the critical size below which dissolution occurs and above which growth occurs.

In the first scenario of pure breakage, milling results in a decrease in the mean particle size over time, and an increase in the number of particles. Increasing the milling intensity leads to a faster reduction in the particle size. In the second scenario of pure Ostwald ripening, crystals above the critical size eventually grow at the expense of the dissolution of crystals below the critical nucleus size (size below which crystals dissolve). As expected, in the absence of nucleation, the number of particles decreases

over time due to dissolution, while the mean particle size and volume increase due to crystal growth. Overall, we note that in the absence of Ostwald ripening we cannot expect generation of supersaturation (and in turn nucleation of a polymorph) and in the absence of particle breakage we cannot expect any growth/dissolution of crystals with large initial mean size (scenario b, dashed lines). Therefore, both these mechanisms were combined and the simulation outcome along with the concept is outlined in Figure 3c. In the combined third scenario, the effects of milling intensity as well as solubility were studied. These simulations were performed under conditions such that, initially, particle breakage is the dominant mechanism which is indeed a true reflection of our experiments (larger initial mean sizes). In the last scenario we observe that the mean particle size and the supersaturation reach a steady-state value, due to the competing effects of simultaneous growth, dissolution, and breakage of crystals. This is in line with the experimental outcome, where particles reach a steady-state particle size with the variations of added solvent in the mill impacting the system's solubility.

The above work sheds significant light on the mechanisms and thermodynamics of milling exemplified with the important system of Ritonavir. The PBEM has shown that both breakage and Ostwald ripening processes are required to explain the milling results. First, large crystals undergo significant size reduction in the ball mill through breakage until Ostwald ripening is triggered which leads to dissolution and growth of particles below and above a critical nucleus size, respectively. The steady-state particle size would be determined by the milling conditions including milling intensity as well as solvent type and concentration (which controls the solubility). Prolonged breakage together with Ostwald ripening establishes a steady-state particle size, with higher solubilities and lower milling intensities leading to larger sizes. The resulting steady-state size along with the supersaturation generated in the mill, will control the thermodynamics at the nanoscale, triggering nucleation and subsequently transformation to the more stable polymorph at the nanoscale.

With molecular modelling and milling, we have shown that not only size but also shape matters in this equilibrium. For RVR, the thermodynamic stability reverses at very small sizes (~40 nm) and for needle crystals, observed in the milled products with SEM. This observation reinforces the necessary role of Ostwald ripening and crystal growth in the mill (leading to needles) beyond the sole role of breakage (leading to blocks). In conditions of abundant solvent and high solubility (large steady state size), RVR-II becomes the most stable phase and is produced readily from RVR-I since abundance of solvent accelerates nucleation and growth. In conditions of lower solvent volumes and low solubility (small size steady state), RVR-I becomes the most stable and can be produced from RVR-II, but it requires long milling times to achieve the steady state. In conditions of no solvent and very low solubility (very small particle size steady state), RVR-A becomes the sole phase. Conditions in between, lead to mixtures of phases. Our results are reproducible, demonstrating that mechanochemistry represents a reliable and consistent tool for polymorph discovery even for conformational phases.

Finally, molecular conformation plays a crucial role in switching thermodynamic stabilities of RVR polymorphs in the mill. As surfaces are generated in milling conditions affording smaller crystals, molecules are stripped off the stabilising intermolecular interactions in the bulk and the conformer energy becomes more dominant. For this reason, at very small particle sizes, the two phases containing the stable trans conformer become thermodynamically favoured (RVR-I and RVR-A). Remarkably, milling forces provide sufficient energy to convert between the trans and the cis conformers in the solid state despite the high energy barrier for the interconversion (>90 kJ mol⁻¹).

The observation that RVR-I readily converts to RVR-II after milling in different solvents raises another important question, that is, whether it is possible that the initial formation of RVR-II during industrial manufacturing in Abbott's laboratories was in fact due to a phase transition of RVR-I caused by mechanical action, which produced the first crystal seeds of RVR-II. Unfortunately, this hypothesis would be difficult to verify. It remains true, however, that RVR-I could have been re-generated from RVR-II by milling with the appropriate conditions, perhaps limiting or avoiding the economic burden suffered by Abbot in the past.

To conclude, the results presented here have shown that breakage and Ostwald ripening processes dictate steady-state crystal size in the mill, that shape as well as size matter in the thermodynamics of nanocrystals and that polymorphs with high energy conformers become thermodynamically unfavoured at the nanoscale. All these point towards a strong prevalence of polymorph stability switches at the nanoscale for conformational polymorphs and to ball-milling as a unique tool to explore polymorphism of complex flexible compounds, especially when guided by computer calculations of particle energies. All these observations have been demonstrated on the important system of Ritonavir.

Experimental Methods

Materials

Ritonavir (RVR, 98%) was purchased from Sigma Aldrich as RVR-I and used without further purification. RVR- II was produced pure by slow evaporation from ethyl acetate solutions. All solvents used were all of analytical grade and used as received. 2-propanol (IPA, $\geq 99.5\%$) and acetonitrile (MeCN, $\geq 99.9\%$) were purchased from Honeywell Research Chemicals, acetone (Ace, $\geq 99.8\%$) from Fisher Scientific UK Ltd and ethyl acetate (EA, 99.8%) from Sigma Aldrich.

Powder X-ray diffraction

Powder X-ray diffraction (PXRD) patterns of all samples were collected on a Bruker D2 Phaser diffractometer equipped with a LYNXEYE detector, using Cu-K α radiation ($\lambda = 1.54 \text{ \AA}$). Intensity data were recorded in the 2θ range of 5-40°. Quantitative phase analysis and crystal size estimates were obtained by Rietveld refinement of collected data with TOPAS v5.^{25,26} The amorphous content of milled samples was determined using corundum ($\alpha\text{-Al}_2\text{O}_3$) as an internal standard.

Solid-State NMR

SSNMR experiments were performed using an Oxford 11.7 T superconducting magnet, and a Bruker AVANCE III HD console, operating at frequencies of 499.69 and 125.65 MHz for ^1H and ^{13}C , respectively. Approximately 100 mg of sample was packed into a zirconia 4 mm rotor with a Kel-F cap. The cross-polarisation magic angle spinning (CP-MAS) with total sideband suppression (TOSS)²⁷ pulse sequence was used for spectral acquisition at a spin rate of 10 kHz using a 4 mm HX magic-angle spinning probe at 25 °C. The magic angle setting was calibrated using KBr. Spectra were obtained using a recycle delay of either 4 seconds (form I and amorphized form I) or 6 seconds (form II and amorphized form II) over 512 (form I and form II) or 800 (amorphized form I and amorphized form II) scans. The ^{13}C chemical shifts were referenced using the high-frequency signal of adamantane ($\delta_{\text{iso}}(^{13}\text{C}) = 38.5 \text{ ppm}$). SPINAL64²⁸ heteronuclear decoupling was used during acquisition with a ^1H nutation frequency of approximately 73.5 kHz. All spectra were processed in Topspin.

FTIR spectroscopy

Solid-state FTIR spectra were recorded using a Thermo Fischer Scientific Nicolet iS50 FTIR Spectrometer.

Scanning electron microscopy (SEM)

SEM images were collected with a Quanta 200 microscope operating under vacuum at 12.5 kV (spot size 2.5). Milled powder samples were dispersed on conductive tape on top of aluminium stubs and coated with 13 nm of platinum.

Ball-mill grinding experiments

Ball-mill grinding experiments were performed with a Retsch MM400 Mixer Mill with screw top 5 ml stainless steel milling jars fitted with a Teflon (PTFE) gasket and 7 mm stainless steel milling balls using a milling frequency of 30 Hz. The duration, as well as the solvent used for each experiment varied as described below. The amount of solvent used is reported using the concentration η (solvent volume/mass of powder, $\mu\text{L}\cdot\text{mg}^{-1}$). The ball milling protocols of Belenguer et al.¹⁵ were followed throughout all experiments and phase composition was determined from PXRD data.

For the initial fast milling screening, neat grinding (NG) and liquid-assisted grinding (LAG) experiments were performed with milling times of 45 minutes starting from 200 mg of either RVR-I or RVR-II. LAG experiments were conducted in presence of 50 μL ($\eta = 0.25 \mu\text{L}\cdot\text{mg}^{-1}$) of various solvents. NG from both forms of RVR resulted in considerable amorphisation of the samples with no polymorphic changes. LAG of the metastable polymorph RVR-I with IPA, MeCN, Ace and EA resulted in the transformation to the bulk-stable RVR-II, whilst LAG with water afforded no polymorphic transformation and little to no amorphisation. LAG of RVR-II with any of the mentioned solvents did

not result in any polymorphic transformation except in the case of water, for which a transition to RVR-I was observed only if longer milling times (> 45 minutes) were used.

We then conducted LAG experiments with IPA for prolonged milling times in order to establish if a steady state for the observed polymorphic transformation of RVR-I to RVR-II had been reached. For different concentrations of IPA, a steady state was attained after just 15 minutes of milling, although the composition of the resulting products varied considerably depending on the concentration η (Figure 1b upper). At $\eta = 0.05 \mu\text{L}\cdot\text{mg}^{-1}$ no conversion to RVR-II was observed; at $\eta = 0.10 \mu\text{L}\cdot\text{mg}^{-1}$ the steady-state product contained a mixture of crystalline RVR-I and RVR-II which was later shown to also contain RVR-A; at $\eta = 0.25 \mu\text{L}\cdot\text{mg}^{-1}$ a full conversion of RVR-I to RVR-II was attained. The time required for the LAG transformation of RVR-II to RVR-I in the presence of water ($\eta = 0.25 \mu\text{L}\cdot\text{mg}^{-1}$) was also studied and was found to be much greater. The full conversion of RVR-II to RVR-I under these conditions typically required about 150 minutes of milling times.

The found milling conditions allowed us to repeatedly interconvert between RVR-I and RVR-II in full one-pot conversion cycles (i.e., LAG of RVR-I with IPA $\eta = 0.25 \mu\text{L}\cdot\text{mg}^{-1}$ for 15 minutes to yield RVR-II; followed by LAG of RVR-II in water $\eta = 0.25 \mu\text{L}\cdot\text{mg}^{-1}$ for 150 minutes to yield RVR-I).

The effect of different solvents and different compositions on the LAG equilibrium milling curves for the polymorphic conversion was also investigated. LAG to steady state should lead to identical milling outcomes independent of the RVR form used initially, only dependent on the milling conditions. Because of that, experiments were performed starting from RVR-I since the milling times to achieve steady state were established to be shorter (45 minutes). Equilibrium milling curves were derived by milling RVR-I with five different solvents (water, IPA, MeCN, Ace, EA) at five different solvent concentrations (from 0.05 to 0.25 $\mu\text{L}\cdot\text{mg}^{-1}$) for 45 minutes (Figure 1c).

Analysis of amorphous ritonavir

The presence of RVR-A was verified by quantitative phase analysis of the milling product of RVR-I with IPA at 0.10 $\mu\text{L}\cdot\text{mg}^{-1}$ for 45 min. These milling conditions were selected for analysis because LAG in IPA had the steepest milling equilibrium curves for the RVR-I to RVR-II transformation compared to other tested solvents (thus the least amount of RVR-A is to be expected). At these LAG conditions, the milling product was determined to be composed of 45% RVR-I, 30% RVR-II and 25% RVR-A.

Further to this, pure RVR-A was produced by NG for prolonged time (120-155 minutes) from RVR-I and RVR-II. A range of characterisation techniques (powder X-ray diffraction, FTIR spectroscopy and ^{13}C solid-state NMR) were used to establish that RVR-A obtained from RVR-I and RVR-II were identical phases.

Computational methods

Calculation of particle energies

The thermodynamic stability of nanosized RVR crystals as a function of particle size (r) and shape (ϕ) was calculated using equation (1).

$$E_{particle}(r, \phi) = (E_{latt-inter} + \Delta E_{intra}) - 0.5 \sum_{(hkl)} x_{(hkl)}(r, \phi) E_{att}^{(hkl)} \quad (1)$$

The first term represents the stabilising (negative) contribution of the bulk crystal structure, and the second term represents the destabilising (positive) contribution of the crystal surfaces. $E_{latt-inter}$ is the intermolecular component of the lattice energy. ΔE_{intra} is the intramolecular conformational energy penalty. $E_{att}^{(hkl)}$ is the attachment energy of the (hkl) facet. The effect of particle size and shape on the particle energy is expressed through the fraction of surface molecules on each (hkl) facet, $x_{(hkl)}(r, \phi)$. Our model assumes crystal particles with idealised shapes and excludes the contribution of surface relaxation and edge energies to the surface energy penalty.

Particle energies were calculated using our Particle Energy Calculator (PEC) code written in Python 3, which utilises modules of the CSD Python API.²⁹ Briefly, PEC takes as an input the crystal structure, the (hkl) attachment energies and an optional user-defined crystal morphology. The main task of PEC is to compute the fraction of surface molecules, $x_{(hkl)}(r, \phi)$, by calculating the volume of convex hulls that are scaled according to the input size. The program can run in two modes: i) generate several crystal morphologies with varying aspect ratios and calculate the particle energy for a user-defined particle size (volume); ii) use a starting morphology to calculate the particle energy as a function of size in a range defined by the user. The results presented in this work were obtained using both modes. A derivation of our model and details about the calculated energy terms and the working principles of the PEC code are presented in the Supplementary Information.

DFT calculations

Periodic DFT calculations of lattice energies ($E_{latt-inter}$) and attachment energies ($E_{att}^{(hkl)}$) were performed with the Vienna ab initio Simulation Package (VASP 5.4.4)³⁰⁻³² with the Perdew-Burke-Ernzerhof³³ functional and Projector Augmented Wave (PAW)^{34,35} potentials release 5.2, using a cut-off energy of 520 eV and applying the Tkatchenko-Scheffler (TS) dispersion correction method.³⁶ The experimental crystal structures with CSD²⁹ refcodes YIGPIO02 (RVR-I) and YIGPIO03 (RVR-II) were used as starting models and their geometries were optimised with tight convergence settings (10^{-8} eV), relaxing both the unit cell parameters and the atomic positions. For the disordered crystal structure of RVR-I (YIGPIO02) the disorder component having the higher stability after optimisation was considered. The intramolecular conformational energy terms of RVR-I and RVR-II, ΔE_{intra} , were computed as described in Beran et al.³⁷

Population balance equation-based model (PBEM)

The evolution of the state of the solid phase and of the liquid phase during milling were described using a PBEM. The model was formulated under the hypothesis that during milling the large crystals undergo significant size reduction through breakage. Upon size reduction, Ostwald ripening is triggered, which leads to dissolution and growth of particles below and above a critical nucleus size (γ_n), respectively. The objective of this model was to verify if prolonged particle breakage and Ostwald ripening can establish a steady state in both particle size and solute concentration.

The PBEM developed in this work accounts for breakage and size-dependent growth and dissolution (Ostwald ripening).^{22,23} The breakage term in our model was simplified and crystal nucleation was assumed negligible. Crystals were assumed to be spherical and described by a single characteristic length (their diameter). Finally, the system was assumed to be composed of particles of a single solid phase. Although we observe polymorphic transformations in the RVR systems studied here, this assumption was made to highlight the interplay between growth, dissolution and breakage mechanisms in reaching a final steady-state of particle size and solute concentration. Note that due to the lack of experimental kinetic data, the PBEM was cast in a non-dimensional form given by

$$\frac{\partial n}{\partial \tau} + \frac{\partial(G^*n)}{\partial y} = B - D \quad (2)$$

where the first term describes the evolution of the number density function n over the dimensionless time τ , the second term accounts for the growth/dissolution of crystal ensemble along their internal coordinate y with a non-dimensional size-dependent velocity G^* (growth/dissolution rate), and the terms on the right-hand side accounts for the birth B and death D of crystals upon breakage. The fundamental equations required to reproduce our model are reported in the Supplementary Information, where both the dimensional and the non-dimensional model are presented. The PBE was solved numerically using a fully discrete high resolution finite volume method.³⁸⁻⁴⁰ Additional details on the solution methodology are provided in the Supplementary Information.

Data availability

All data that support the findings of this study are available within the paper and its Supplementary Information, or from the corresponding authors upon request.

Code availability

The PEC code is available upon request.

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P.S. performed the molecular modelling simulations and wrote the PEC code; S.E.W. performed the milling experiments and the quantitative phase analysis of PXRD data with significant contributions from G.I.L.; Population balance equation modelling was performed by P.N. with key discussions and contributions from A.K.R.; W.K. performed milling experiments and collected the SEM images. SSNMR experiments were performed by C.L.E. and P.H. A.J.C.C. led the work, discussions and writing of the article and developed the research plan. All authors were involved in lengthy discussions and contributed to the writing of the article.

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Ethics declarations

Authors have nothing to declare.