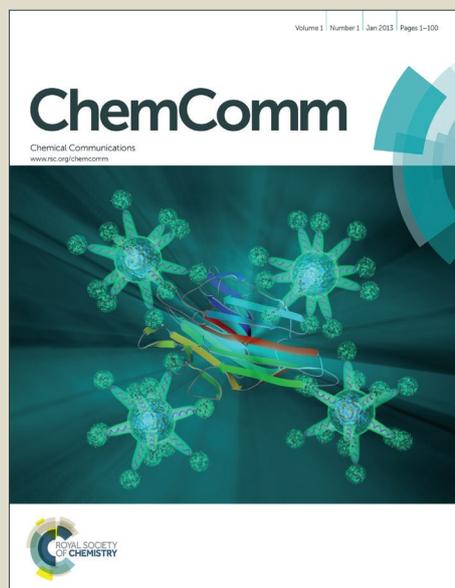


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How cocrystals of weakly basic drugs and acidic cofomers might modulate solubility and stability

Received 00th January 20xx,
Accepted 00th January 20xx

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DOI: 10.1039/x0xx00000x

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Cocrystals of a weakly basic drug (nevirapine) with acidic cofomers are shown to alter the solubility dependence on pH, and to exhibit a pH_{max} above which a less soluble cocrystal becomes more soluble than the drug. The cocrystal solubility advantage can be dialed up or down by solution pH.

Cocrystals can increase drug solubilities by orders of magnitude, yet such enhancement can be thwarted by the extent of ionization¹⁻³ and solubilization⁴⁻⁷ of cocrystal components. For this reason cocrystal solubility only makes sense in light of solution conditions, such as pH (ionization) and additives (solubilizing agents). The present work shows (1) how solubility and thermodynamic stability of cocrystals can change with solution pH, and (2) best and rapid approaches to characterize such behavior.

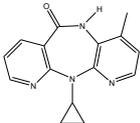
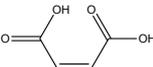
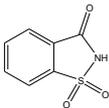
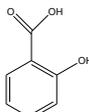
NVP is a non-nucleoside reverse transcriptase inhibitor (NNRTI)⁸ used in the treatment of HIV-1 infections. NVP is a lipophilic molecule that exhibits poor oral bioavailability due to dissolution rate limited absorption.⁹ Formation of NVP cocrystals as a means to enhance aqueous solubilities relative to the pure crystalline drug was recently investigated by Caira et al.¹⁰ Several NVP cocrystals with acidic cofomers, maleic acid (MLE), saccharin (SAC) and salicylic acid (SLC), among others, were discovered. The authors of this fine publication observed that dissolution studies failed to reveal the expected increases in cocrystal solubility, based on high cofomer to drug solubilities; a trend that our group has confirmed for other cocrystals.¹¹ This motivated us to investigate the reasons for such unexpected findings, and gain a deeper understanding of how these cocrystals work. Chemical

structures and pK_a values of drug and cofomers in the cocrystals we studied are summarized in Table 1.

The influence of pH on cocrystal solubility has not generally been recognized and very few articles report the pH associated with cocrystal dissolution and solubility measurements^{1, 3, 6, 7, 12}. Cocrystals of a basic drug and an acidic cofomer will encounter solution conditions under which the drug and/or cofomer may be ionized. Since cocrystal solubility is primarily influenced by the sum of all the cocrystal constituent species in solution (ionized and nonionized, in this case), pH is expected to be a crucial factor in determining such cocrystal solubilities.

Fig. 1 shows the solubility of NVP and its cocrystals as a function of pH. These results demonstrate that cocrystals change the solubility vs pH curve from an exponential decrease leading to a constant low value for NVP, to a “U shaped” curve with exponentially decreasing and increasing solubilities for its cocrystals. NVP is highly soluble at $pH < 3$, but its solubility decreases by about 2 orders of magnitude

Table 1. Chemical structures and pK_a values of drug and cofomers.

Compound	Chemical structure	pK_a
Nevirapine		2.8 ¹³
Maleic acid		1.9 and 6.6 ¹⁴
Saccharin		1.6 ¹⁵
Salicylic acid		3.0 ¹³

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Supplementary Information (ESI) available: Experimental details and solubility data. See DOI: 10.1039/x0xx00000x

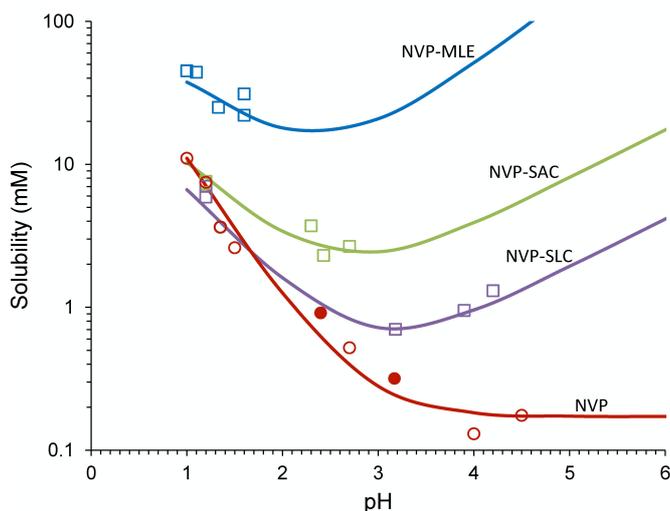


Fig. 1 Solubility of the basic drug NVP and its cocrystals with acidic cofomers: (1:1) cocrystal NVP-MLE, and (2:1) NVP-SAC and NVP-SLC as a function of pH. Symbols represent solubilities determined from solutions saturated with NVP and/or cocrystal at 25°C. pH values correspond to equilibrium pH. The pH value at the intersection of the drug and cocrystal (NVP-SAC and NVP-SLC) solubility curves corresponds to pH_{max} or transition point above which a less soluble cocrystal becomes more soluble than drug. Curves were calculated from cocrystal and drug solubility-pH dependence according to equations 1 and 2 and parameter values presented in the text and in table 2. Symbols represent: NVP solubility (NVP hydrate-open circles, NVP anhydrous-filled circles) and cocrystal solubilities from eutectic points (squares).

(pH 1 to 4) to a constant low solubility value of approximately 0.2mM at $pH > 4$. Cocrystals on the other hand, show a solubility decrease (pH 1 to 3) and a solubility increase at $pH > 3$. Some cocrystals also exhibit a transition point or pH_{max} at the intersection of the drug and cocrystal solubility curves. This means that cocrystals will exhibit higher solubilities than drug at $pH > pH_{max}$.

Solubility-pH relationships are well recognized for acids and bases, and their corresponding salts¹⁶⁻¹⁸. NVP is a weak base and its solubility as a function of pH is described by

$$S_{drug} = S_{drug,0} (1 + 10^{pK_{a,drug} - pH}) \quad (1)$$

where $S_{drug,0}$ represents the solubility of NVP under nonionizing conditions, 0.172mM at 25°C. K_a represents the ionization constant of NVP conjugate acid. This equation shows that the simultaneous measurement of solubility and pH allows for calculation of solubility at other pH values.

Cocrystal solubility is the sum of all the cocrystal component species in solution that are in equilibrium with the cocrystal. Theoretical solubility-pH relationships for NVP cocrystals can therefore be derived, by considering the two relevant equilibria from which the concentration of cocrystal component species can be obtained. The first is the cocrystal dissolution/dissociation equilibrium defined by a solubility product, K_{sp} , and the second involves the ionization of drug and cofomer characterized by their respective ionization

constants, K_a . Derivation of cocrystal solubility equations is presented in the ESI†. The solubility expression is then obtained by considering the mass balance of each cocrystal component (ionized and unionized species) to give

$$S_{cc}^{1:1} = \sqrt{K_{sp} (1 + 10^{pK_{a,D} - pH}) \times \sqrt{(1 + 10^{pH - pK_{a1,CF}} + 10^{2pH - pK_{a1,CF} - pK_{a2,CF}})}} \quad (2)$$

for the 1:1 cocrystals with diprotic acidic cofomers. Subscripts D and CF represent drug and cofomer. For 2:1 cocrystals with monoprotic acidic cofomers, the solubility equation becomes

$$S_{cc}^{2:1} = 2 \sqrt[3]{\frac{K_{sp}}{4} (1 + 10^{pK_{a,D} - pH})^2 (1 + 10^{pH - pK_{a1,CF}})} \quad (3)$$

Cocrystal solubility in the above equations is expressed in terms of moles of drug, unless otherwise indicated.

Excellent agreement between measured and predicted cocrystal solubilities in Fig. 1, demonstrates that these equations predict how pH might influence cocrystal solubility from knowledge of cocrystal K_{sp} , and K_a values of its components. Cocrystal K_{sp} values in this work were determined from cocrystal solubility measurements under equilibrium conditions at the cocrystal/NVP hydrate eutectic points unless otherwise indicated.

Caira et al.¹⁰ characterized cocrystal solubilities from cocrystal dissolution in water. Our solubility studies suggest that the unexpectedly moderate cocrystal solubility increases (S_{cc}/S_{drug}) reported by Caira et al.¹⁰ (Table 2) are due to the sensitivity of cocrystal solubility on pH, and to possible transformation of cocrystals to NVP. As shown in Fig. 1, NVP cocrystal solubilities can: (1) vary by orders of magnitude with pH, and (2) approach drug solubility as pH approaches pH_{max} .

Table 2 Nevirapine cocrystals: K_{sp} , pH_{max} , and S_{cc}/S_{drug} .

Cocrystal	K_{sp} ^a (M ² or M ³) ^b	pH_{max} ^c	S_{cc}/S_{drug} ^d pH 1 to 5	S_{cc}/S_{drug} ^e pH ?
NVP-MLE (1:1)	1.96×10^{-5}	none	3.4–906	5.3
NVP-SAC (2:1)	1.05×10^{-10}	1.1	0.9–47	1.4
NVP-SLC (2:1)	3.63×10^{-11}	1.7	0.6–11	1.1

(a) Calculated from equilibrium solubility measured at cocrystal/drug eutectic points at 25°C described in ESI†. (b) Units of M² for 1:1 and M³ for 2:1 cocrystals. (c) Obtained from the intercept of drug and cocrystal solubility curves in Fig. 1. (d) Obtained from equilibrium solubility calculation, S vs pH curves in Fig. 1. (e) From Caira et al.,¹⁰ obtained from cocrystal dissolution in water, pH unknown, and NVP solubility in water (0.36mM) at 37°C. The influence of temperature on S_{cc}/S_{drug} is expected to be small compared to the influence of pH. S_{drug} hydrate increases by about 2 fold between 25 and 37°C¹⁹ and the change in S_{cc}/S_{drug} may be even smaller if at all.

The S_{cc}/S_{drug} reported by Caira et al.¹⁰ is highest for the NVP-MLE with a value around 5. We found that this cocrystal is the most soluble of the three cocrystals we studied and does not have a pH_{max} . In addition, S_{cc}/S_{drug} is hugely dependent on pH with a value of 3 at pH 1, 20 at pH 2, and 900 at pH 5. Having the most soluble coformer at the highest molar ratio (1:1) appears to contribute to the high solubility of this cocrystal.

The S_{cc}/S_{drug} values for NVP-SAC and NVP-SLC cocrystals were lower than for NVP-MLE, consistent with our findings. Furthermore, the reported¹⁰ S_{cc}/S_{drug} values were around 1, suggesting the proximity of solution pH to pH_{max} . We discovered that these cocrystals have pH_{max} values of 1.1 and 1.7 where $S_{cc}/S_{drug} = 1$. Without knowledge of the pH or pH_{max} , the increase in solubility that these cocrystals impart could be missed. We observed that dissolution of these three cocrystals lowered solution pH as the acidic coformer concentrations increased (as shown by the decrease in initial pH as solutions reached equilibrium). For these reasons it is essential to measure the pH associated with cocrystal solubility or dissolution studies.

It is important to note that S_{cc} values determined by kinetic methods are generally lower than those measured by equilibrium methods used in our work, as a result of the cocrystal conversion to the constituent drug during dissolution. S_{cc}/S_{drug} values determined from eutectic point measurements are equilibrium values and provide a supersaturation index. This means that the huge increase in S_{cc}/S_{drug} with pH translates to very high supersaturation with respect to the less soluble drug, leading to more favorable and faster conversions to drug. While the higher S_{cc}/S_{drug} values may not be experimentally reached they give insight as to the conversion rates that a cocrystal might experience and inform dissolution and cocrystal formulation approaches.

We have previously demonstrated the importance of the eutectic constant, K_{eu} , as a key indicator of cocrystal to drug solubility, and as an experimentally accessible equilibrium state regardless of the cocrystal solubility relative to pure components.^{2,20} K_{eu} is defined as

$$K_{eu} \equiv \frac{[coformer]_{eu,total}}{[drug]_{eu,total}} \quad (4)$$

at the eutectic point where drug and cocrystal are in equilibrium with solution. The terms in brackets represent concentrations. Subscripts eu and total are analytical concentrations (ionized+nonionized) at the eutectic point.

K_{eu} values above cocrystal stoichiometric ratio, indicate that the cocrystal is more soluble than the drug, and the opposite for cocrystals that are less soluble than drug. For the purpose of initially evaluating the NVP cocrystal to drug solubility and stability, we determined K_{eu} values from measured eutectic concentrations of coformer and drug as shown in Fig. 2. Studies were done at 25°C and pH values between 1 and 4 (Fig. 2). Higher pH values could not be reached due to buffering by the acidic cofomers.

For the 1:1 NVP-MLE cocrystal, $K_{eu} > 1$ at all pH values studied indicating that the cocrystal is more soluble than the

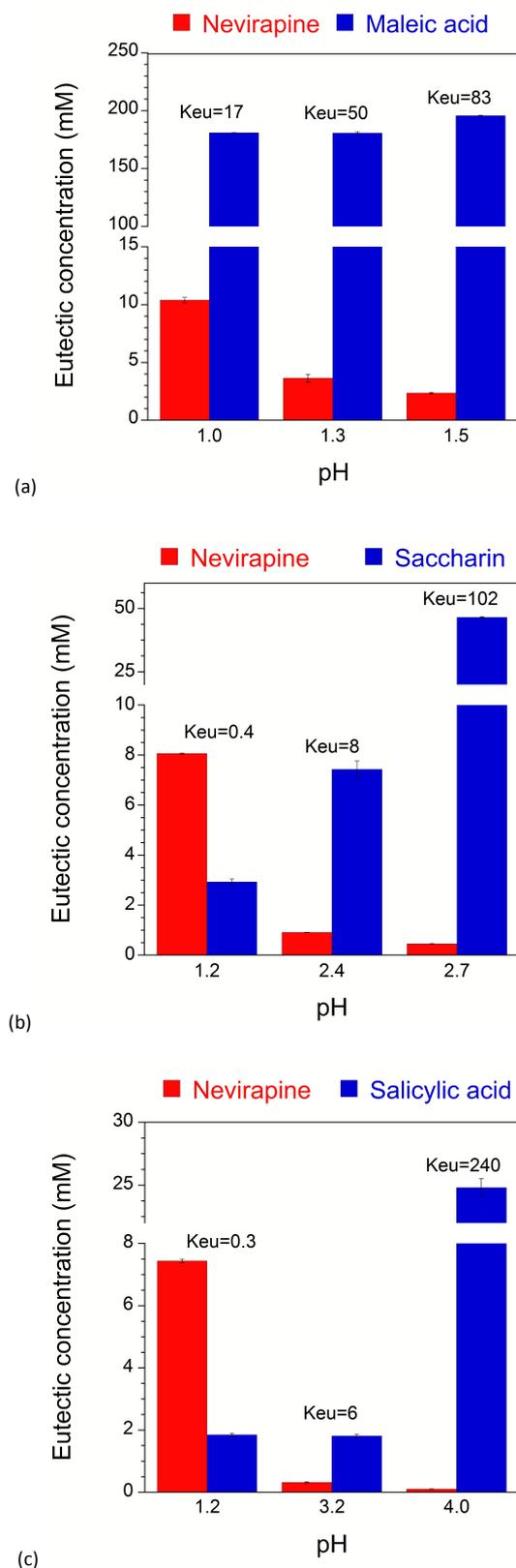


Fig. 2 Eutectic concentrations of drug and coformer at different pH values are key indicators of cocrystal thermodynamic stability relative to drug. a) NVP-MLE (1:1), b) NVP-SAC (2:1), and c) NVP-SLC (2:1). NVP-MLE cocrystal is more soluble than NVP as suggested by $K_{eu} > 1$, $[coformer]_{eu} > [drug]_{eu}$. SAC and SLC cocrystals have a reversal in this trend as pH increases, $K_{eu} < \text{or} > 0.5$, indicating a transition point at pH_{max} . pH values represent equilibrium values and are generally lower than initial pH.

drug since $S_{cc}/S_{drug} > 1$. This cocrystal does not have a pH_{max} where $K_{eu} = 1$.

For 2:1 cocrystals a pH_{max} occurs at $K_{eu} = 0.5$. Both NVP-SAC and NVP-SLC cocrystals show $K_{eu} < 0.5$ at pH 1.2 while K_{eu} is > 0.5 for NVP-SAC at pH values ≥ 2.4 , and for NVP-SLC at pH values ≥ 3.2 , demonstrating that there is a pH_{max} for both cocrystals. These results are in excellent agreement with those predicted from the solubility curves in Fig. 1. Small deviations in pH_{max} for the NVP-SAC cocrystal where $K_{eu} = 0.4$ suggests a pH_{max} slightly higher than 1.1, estimated from Fig. 1. This small difference in pH_{max} values is a consequence of the variability between predicted (Eq.1) and experimentally determined NVP solubility as a function of pH (shown in ESI†).

We have previously demonstrated that S_{cc}/S_{drug} can be estimated from K_{eu} according to^{2, 20}:

$$K_{eu}^{1:1} = \left(\frac{S_{cocrystal}}{S_{drug}} \right)^2 \quad (5)$$

$$K_{eu}^{2:1} = 0.5 \left(\frac{S_{cocrystal}}{S_{drug}} \right)^3 \quad (6)$$

S_{cc} and S_{drug} were experimentally determined from measured eutectic concentrations (Fig. 2) for each cocrystal and respective pH, as described in ESI†. The experimental and predicted K_{eu} dependence on S_{cc}/S_{drug} is presented in Fig. 3. The results show excellent agreement between observed and predicted behavior according to equations 5 and 6 for 1:1 and 2:1 cocrystals, respectively.

In conclusion, cocrystal solubility and its advantage over drug can be dialed up or down by solution pH. Cocrystal and drug solubilities without measurement of the corresponding pH will fail to provide meaningful insight about how cocrystals dissolve, and in some cases miss their ability to enhance and

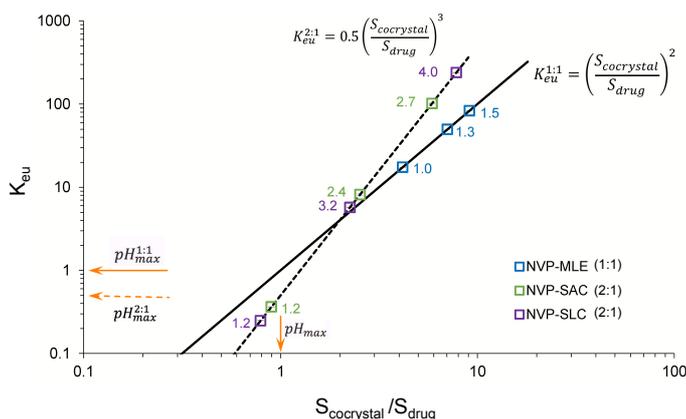


Fig. 3 Predicted and experimental values of K_{eu} and cocrystal solubility advantage (S_{cc}/S_{drug}) for 1:1 NVP-MLE and 2:1 NVP-SAC and NVP-SLC cocrystals. K_{eu} is a key indicator of S_{cc}/S_{drug} . K_{eu} dependence on pH reveals the cocrystal pH_{max} as well as the cocrystal increase in solubility over drug as pH increases. At pH_{max} , $K_{eu} = 1$ for 1:1 cocrystals and $K_{eu} = 0.5$ for 2:1 cocrystals. Log axes are used due to the large range of values. Symbols represent experimental values. Numbers next to data points indicate pH at eutectic point or equilibrium pH. Lines were generated according to equations 5 and 6. Solid lines represent 1:1 cocrystals and dashed lines 2:1 cocrystals.

modulate solubility. We have also demonstrated that eutectic constants are key indicators of cocrystal stability and solubility. Their measurement requires small amount of materials and time for a solution to reach equilibrium with. K_{eu} also provides a supersaturation index, which is the driving force for cocrystal transformation to the less soluble drug. A subsequent publication will address the influence of the supersaturation index on cocrystal dissolution.

We gratefully acknowledge partial support from CAPES-Brazil/PVE-Program. GK also acknowledges CAPES-Brazil/PDSE scholarship (process 7537/13-1). NRH and GK are grateful for partial support from the National Institute of General Medical Sciences of the National Institutes of Health (NIH) under award number R01GM107146. The content is solely the responsibility of the authors and does not necessarily represent the official views of the NIH.

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