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Predicting the Formation and Stability of Amorphous Small Molecule Binary Mixtures from Computationally Determined Flory−Huggins Interaction Parameter and Phase Diagram

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Abstract: The Flory−Huggins interaction parameter has been shown to be useful in predicting the thermodynamic miscibility of a polymer and a small molecule in a binary mixture. In the present paper, this concept was extended and evaluated to determine whether or not the Flory−Huggins interaction parameter can be applied to small molecule binary mixtures and if this parameter can predict the phase stability of such amorphous binary mixtures. This study was based on the assumption that a thermodynamically miscible binary system is stable and cannot crystallize, and that phase separation is essential before the individual components can crystallize. The stabilization of a binary system is thought to derive from molecular interactions between components in a solid dispersion, which are characterized by the Flory−Huggins interaction parameter. Based on DSC experiments, drug molecules (39) in the present study were classified into three different categories according to their crystallization tendency; i.e., highly crystallizing, moderately crystallizing and noncrystallizing compounds. The Flory−Huggins interaction parameter was systematically calculated for each drug pair. The validity of this approach was empirically verified by hot-stage polarized light microscopy. If both compounds in the pair belonged to the category of highly crystallizing compound, the Flory−Huggins interaction predicted an amorphous or crystalline phase with approximately 88% (23 out of 26) confidence. If one or both compounds of the pair were either moderately crystallizing or noncrystallizing compounds, the binary mixture remained in the amorphous phase during the cooling phase regardless of the interaction parameter. The Flory−Huggins interaction parameter was found to be a reasonably good indicator for predicting the phase stability of small molecule binary mixtures. The method described can enable fast screening of the potential stabilizers needed to produce a stable amorphous binary mixture.

Keywords: Amorphous; miscibility; stabilization; Flory−Huggins; solid dispersion

Introduction

Poor solubility or a slow dissolution rate of a drug often leads to low bioavailability and, hence, may complicate the exploitation of otherwise potentially new drug candidates. One solution to this problem may be achieved by converting a poorly soluble crystalline drug compound into an amorphous form. In general, amorphous solids have a higher apparent solubility and faster dissolution rates than their crystalline counterparts[1−4] and, hence, may provide formulation alternatives to enhance bioavailability.[5] However, thermodynamic instability is the main reason why amorphous drugs have not been more widely used to improve bioavailability, especially in commercial products, and there are several methods that have been developed to circumvent this problem. Some of these methods utilize crystalline-inhibiting additives that stabilize amorphous structures,[6−9] although most of these methods are not universally applicable and only work for specific drug formulations. In addition, there is a lack of reliable or systematic methods that can both quickly and reliably screen a large number of excipients in order to identify potentially useful candidates that could possibly inhibit crystallization or stop the nucleation already initiated.

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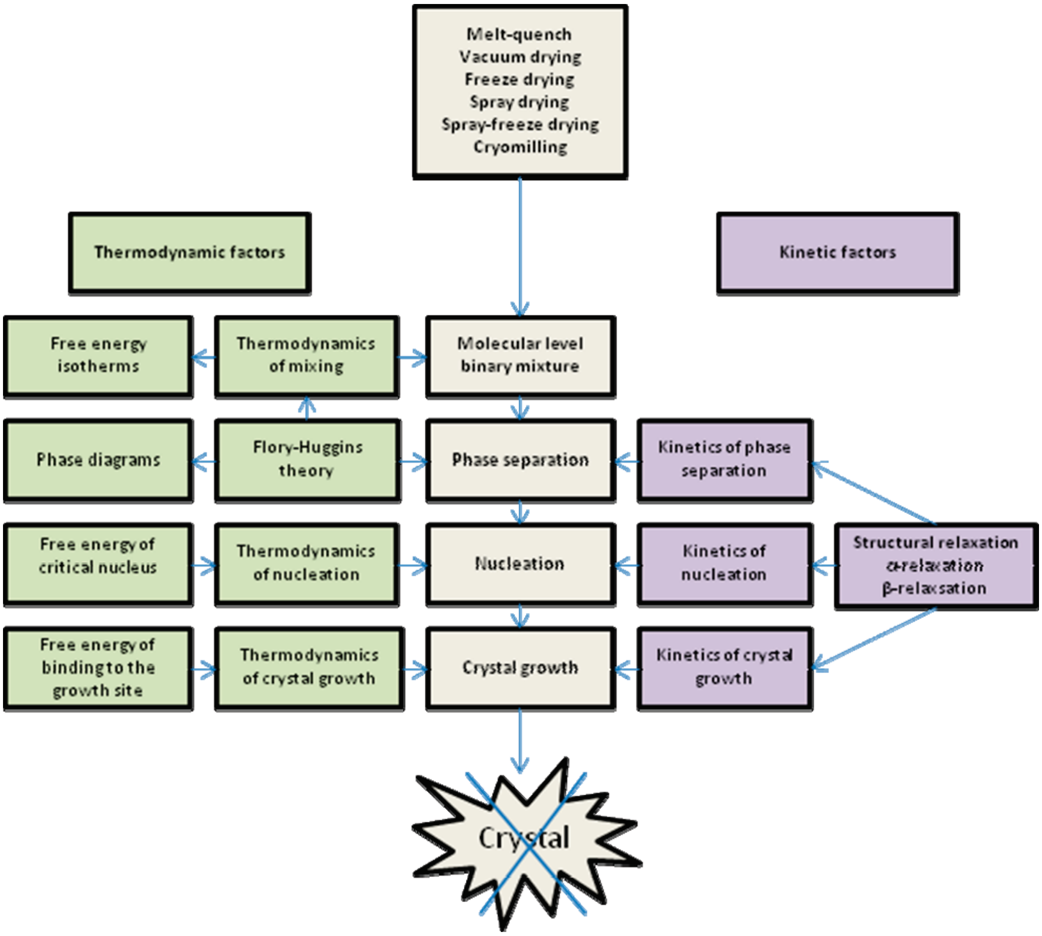

Figure 1. Schematic pathway to describe the crystallization of an amorphous binary system. By controlling factors that influence each step, crystallization can be prevented and the amorphous structure stabilized.

Our research strategy for stabilizing amorphous small molecule mixtures has been based on an understanding of how to control/stabilize, thermodynamically and kinetically, high energetic amorphous binary systems. A flowchart was devised to guide in the selection of thermodynamically based excipients for the stabilization of various amorphous solids and describe the crystallization pathway of an amorphous binary system (Figure 1).

The first step involves preparing a molecule level mixture of the amorphous binary system with an appropriate method. At this point, it should be emphasized that a totally random arrangement of drug and excipient molecules is a prerequisite for the successful stabilization of an amorphous binary system. In the second step, we assume that a thermodynamically miscible binary system is stable and cannot crystallize, and requires phase separation to occur before the individual components can crystallize. In other words, an excipient is selected that forms a thermodynamically miscible mixture and a stable binary system with a drug compound, which then enables the formation of a stable amorphous structure. Finally, the last two steps (nucleation and crystal growth)

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are the worst-case scenarios that should be avoided if the first two steps have been successful. However, if the system does progress to nucleation and crystal growth, we present ways to minimize crystallization.

In technical terms, this model does not take into account whether or not crystallization arises because of phase separation or cocrystallization. It is possible that crystallization initiates the formation of cocrystals, which might be desirable if the physicochemical properties of the cocrystals are better than the pure components. However, the primary goal of this research was to stabilize high energetic amorphous structures and, hence, the accuracy of the described method was verified on the grounds of crystallized or noncrystallized binary system, regardless of whether or not the crystals were cocrystalline or pure individual components.

In general, the problem can be described by various thermodynamic and reaction kinetic models (i.e.; Arrhenius, classical nucleation theory, Flory−Huggins for miscibility). These transformational stages are governed by both thermodynamic and kinetic factors that must be taken into account for the successful development of a stable amorphous binary system. The focus of this paper is on the second step of our pathway model. Emphasis will be placed on how to screen rapidly, and with great validity, the miscibility of two components and how to distinguish between stable (desired) and phase separated (undesired) systems in vitro. No long-term stability studies were included in the present studies. However, the long-term stability and additional steps in the pathway model will be studied in subsequent papers.

Background for Miscibility of a Two-Component System. According to several studies, there are different ways to determine miscibility; e.g., by calculating the solubility parameters[10−14] and partition coefficients,[15] using DSC methods to define the crossing point of the dissolution end point T_{end} and T_{g}[16] or defining the correlation by the Gordon−Taylor equation for calculated glass transition temperatures and comparing with observed glass transition temperatures.[17−20] The key element in some of these methods is evaluating the miscibility of components by deriving the Flory−Huggins interaction parameter as a measure of miscibility. The methods used to estimate interaction parameters include melting point depression and the determination of solubility parameters using group contribution theory. In our study, these methods are extended to include a computational method that has been used to define the interaction parameters of small molecular weight substances, instead of polymers, as crystallization inhibitors.

The aim of the present study was to prevent the crystallization of amorphous structures by using computational methods to identify potential stabilizers. Hence, the very first step in our model was the preparation of a well-mixed amorphous binary system. For the successful stabilization of an amorphous binary system, it is a prerequisite that arrangement of drug and excipient molecules is totally random. For mixing an excipient with a drug at a constant temperature, T, the Gibbs free energy change of mixing, ΔG_{mix}, can be defined by eq 1,

$$\Delta G_{\mathrm{mix}} = \Delta H_{\mathrm{mix}} - T\Delta S_{\mathrm{mix}} \tag{1}$$

in which ΔH_{mix} and ΔS_{mix} are enthalpy and entropy of mixing, respectively.[21] For spontaneous mixing, ΔG_{mix} must be negative and ΔS_{mix} is always positive (i.e., the contribution for ΔG_{mix} is negative, since the $-T\Delta S_{\mathrm{mix}}$ term is negative and greater than ΔH_{mix}) because the measure of randomness increases as the mixing of the two components occurs. Thus, for negative ΔG_{mix}, ΔH_{mix} has to be negative or only slightly positive. The Flory−Huggins theory is a well-known thermodynamic equation for mixing and phase separation,[21] and defines the change in the free energy of mixing as follows (eq 2):

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$$\Delta G_{\text{mix}} = RT(n_{\text{A}} \ln \phi_{\text{A}} + n_{\text{B}} \ln \phi_{\text{B}} + \chi_{\text{AB}} n_{\text{A}} \phi_{\text{B}}) \quad (2)$$

where R and T are the gas constant and temperature, respectively, n_i and ϕ_i are the number of moles and volume fractions of components A and B, respectively, and χ_{AB} is the Flory−Huggins interaction parameter (also referred to as the solubility parameter). The two first terms account for combinatorial entropy contributions, and the third term is an enthalpic contribution. As stated above, the entropic contribution is always negative (favorable), but the enthalpic contribution can be ether positive or negative. In the enthalpy term (eq 2), the interaction parameter defines the compatibility of mixing two different molecules. Thus, negative or slightly positive χ_{AB} values will guarantee the miscibility of a two-component system. In other words, adhesive forces (drug−excipient) have to be greater than cohesive forces (drug−drug and/or excipient−excipient forces).

The interaction parameter χ_{AB} can be determined experimentally from melting point depression, solubility, or ellipsometric measurements, and by several scattering methods or solubility parameters (Hildebrand and Hansen solubility parameters), which are related to the square root of cohesive energy density, molar energy or the heat of vaporization of a pure liquid. Hildebrand and Hansen solubility parameters are determined by using group contribution methods that involve the summation of individual functional group contributions of both cohesive energy density (CED) and volume to the solubility parameter. The Hildebrand solubility parameter is a single value, although the Hansen solubility parameter takes into account the different types of interactions to provide partial solubility parameters.

CED can be determined by utilizing modern computational methods (i.e., molecular dynamic simulations). In an atomistic simulation, the cohesive energy is defined as the increase in energy per mole of a material when all intermolecular forces are eliminated. The CED corresponds to the cohesive energy per unit volume, i.e., the energy difference between a molecule in vacuum state and in the bulk amorphous state within a unit volume (eq 3):

$$\delta_{\text{hil}} = (\text{cohesive energy density})^{1/2} = ((E_{\text{vacuum}} - E_{\text{bulk}})/V)^{1/2} \quad (3)$$

where δ_{hil} is the Hildebrand solubility parameter, E_{vacuum} and E_{bulk} are the energy values in vacuum and in bulk, respectively, and V is the volume of the periodic cell. Furthermore, the energy for mixing components can be determined from the cohesive energy densities (eq 4):

$$\Delta E_{\text{mix}} = \phi_{\text{A}}(E_{\text{coh}}/V)_{\text{A}} + \phi_{\text{B}}(E_{\text{coh}}/V)_{\text{B}} - (E_{\text{coh}}/V)_{\text{mix}} \quad (4)$$

where ϕ_{A} and ϕ_{B} are volume fractions of components A and B in a mixed system, respectively. Finally, the Flory−Huggins interaction parameter χ_{AB} can be determined from the mixing energy (eq 5).

$$\chi_{\text{AB}} = \Delta E_{\text{mix}}/RT \quad (5)$$

In general, a negative value for the interaction parameter at a certain temperature indicates that two different molecules have a more favorable interaction with each other in preference to interacting with similar molecules, and would most probably exist in one phase. Large positive values indicate that the molecules will preferably interact with similar molecules and that phase separation will most likely occur. The disadvantage of calculating the Flory−Huggins interaction parameter χ_{AB} from molecular dynamic simulations is that this usually requires quite a long simulation to achieve an equilibrium state. In addition, the molecular dynamic simulation requires careful selection of the force field, charge model and simulation parameters. This means that it is currently almost impossible to screen large compound sets due to the long simulation times involved.

In the present study, an alternative approach was used to determine the Flory−Huggins interaction parameter χ_{AB}, by using the Material Studio Blends module (Accelrys Inc., San Diego, CA).[22] The original Flory−Huggins theory is based on a simple lattice model and has several limitations. It does not take into account the volume change from mixing (constant lattice size/volume) and is composition independent of χ_{AB}. In the Blends module, extended Flory−Huggins theories have been implemented in the present study to overcome the above-mentioned shortcomings. The Blends module calculates the χ_{AB} parameter for molecules in a binary mixture by combining the modified Flory−Huggins model and molecular modeling techniques. The extensions to the Flory−Huggins theory, where molecules are arranged on a regular lattice, consider those molecules that can be arranged irregularly in the lattice (i.e., the off-lattice model), and that incorporate an explicit temperature dependence on the interaction parameter.[23] The temperature-dependent χ_{AB} parameter is calculated explicitly by generating a large number of pair configurations and calculating binding energies by Monte Carlo simulation, followed by temperature averaging of the results by using the Boltzmann distribution factor. Arranging molecules off-lattice relates to the coordination number, Z, which is the number of molecules A in the role of screen (s), that can be packed around a single base (b) molecule B. By calculating an average of temperature-dependent binding energies and coordination numbers of all possible pair configurations of molecules A and B, the energy of mixing at temperature T can be calculated by using eq 6:

$$\Delta E_{\text{mix}} = {}^{1}/_{2}[Z_{\text{bs}}(E_{\text{bs}})_T + Z_{\text{sb}}(E_{\text{sb}})_T - Z_{\text{bb}}(E_{\text{bb}})_T - Z_{\text{ss}}(E_{\text{ss}})_T] \quad (6)$$

where Z is a coordination number, E is the binding energy, and lower case s and b are base and screen molecules, respectively.

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Table 1. The Drug Compounds Evaluated[a]

drug compound	mp (°C)	T_g (°C)	crystallization temp (°C)	supplier	purity
nabumetone[b]	80.79 ± 0.07		51.53 ± 2.57	Sigma-Aldrich	≥98%
benzocaine[b]	89.72 ± 0.01		54.67 ± 12.64	Sigma-Aldrich	99%
chlorpropamide[b]	123.95 ± 0.11		80.57 ± 0.54	Sigma-Aldrich	97%
tolbutamide[b]	128.50 ± 0.05		63.1 ± 18.43	Sigma-Aldrich	99.8%
salicylamide[b]	140.31 ± 0.01		123.92 ± 3.17	Sigma-Aldrich	99%
naproxen[b]	155.33 ± 0.08		121.75 ± 2.20	Sigma-Aldrich	100.2%
salicylic acid[b]	158.82 ± 0.01		107.56 ± 1.00	Sigma-Aldrich	99%
thiosalicylic acid[b]	163.84 ± 0.43		146.18 ± 0.11	Sigma-Aldrich	95%
sulfanilamide[b]	164.18 ± 0.08		127.94 ± 1.40	Sigma-Aldrich	min 99%
sulfamethoxazole[b]	169.19 ± 0.14		147.40 ± 3.54	Sigma-Aldrich	min 98%
fenbufen[b]	185.77 ± 0.15		152.69 ± 0.30	Sigma-Aldrich	>99%
pyrazinecarboxamide[b]	189.05 ± 0.06		177.73 ± 1.62	Sigma-Aldrich	98%
piroxicam[b]	200.36 ± 0.04		163.19 ± 6.99	Sigma-Aldrich	≥98%
diflunisal[b]	212.29 ± 0.26		201.38 ± 1.50	Sigma-Aldrich	98.4%
tolfenamic acid[b]	212.15 ± 0.13		163.91 ± 10.01	Sigma-Aldrich	99.6%
saccharin[b]	227.94 ± 0.02		160.40 ± 59.07	Sigma-Aldrich	≥99.0%
mefenamic acid[b]	230.32 ± 0.08		142.36 ± 9.85	Sigma-Aldrich	98%
sulfamerazine[b]	236.57 ± 0.07		174.05 ± 8.02	Sigma-Aldrich	99%
primidone[b]	283.16 ± 0.14		192.77 ± 5.37	Sigma-Aldrich	min 98%
flurbiprofen[c]	113.97 ± 0.21	−4.31 ± 0.08	41.70 ± 0.12	Sigma-Aldrich	min 99%
flutamide[c]	111.84 ± 0.03	2.08 ± 0.21	41.07 ± 0.46	Sigma-Aldrich	99%
acetylsalicylic acid[c]	136.09 ± 0.35	−26.92 ± 0.22	29.23 ± 1.03	Sigma-Aldrich	>99%
nimesulide[c]	148.64 ± 0.05	21.60 ± 0.32	78.98 ± 0.66	Sigma-Aldrich	98%
paracetamol[c]	167.82 ± 0.34	25.01 ± 0.30	75.58 ± 1.14	Orion Pharma	99.0−101.0%
sulfadimidine[c]	196.80 ± 0.07	78.12 ± 0.25	144.13 ± 8.84	Orion Pharma	99.0−101.0%
ibuprofen[d]	74.76 ± 0.33	−38.71 ± 0.35		Sigma-Aldrich	≥98%
ketoprofen[d]	94.60 ± 0.29	−1.97 ± 0.15		Sigma-Aldrich	≥98%
perphenazine[d]	93.97 ± 0.23	15.31 ± 0.23		Sigma-Aldrich	min 99%
captopril[d]	106.19 ± 0.05	1.67 ± 0.16		Sigma-Aldrich	≥98%
nizatidine[d]	132.14 ± 0.29	11.30 ± 0.08		Sigma-Aldrich	100%
cimetidine[d]	139.41 ± 0.17	46.52 ± 0.17		Sigma-Aldrich	98%
clotrimazole[d]	142.97 ± 0.09	29.77 ± 0.18		Sigma-Aldrich	98%
pyridoxine[d]	157.25 ± 0.09	66.26 ± 2.49		Sigma-Aldrich	≥98%
famotidine[d]	162.40 ± 0.41	74.51 ± 0.22		Sigma-Aldrich	99%
furosemide	degraded			Sigma-Aldrich	98%
sulfadiazine	degraded			Sigma-Aldrich	99%
acetazoleamide	degraded			Sigma-Aldrich	≥99%
nitrofurantoin	degraded			Sigma-Aldrich	98%
chlorthiazide	degraded			Sigma-Aldrich	≥98%

[a] Melting point and crystallization temperatures were determined as onset values and glass transition temperatures as midpoint values ($n = 3$ for each drug). [b] Highly crystallizing compound. [c] Moderately crystallizing compound. [d] Noncrystallizing compound.

These extensions allow the determination of the temperature dependent interaction parameter, temperature and composition dependent free energy of mixing, the phase diagram, the critical point as well as the coexisting and stability curves of the binary system. One major advantage of this procedure is that the generation of paired configurations via Monte Carlo simulations and single point energy calculations is much faster than by molecular dynamic simulations. This makes it possible to screen much larger compound sets for a certain drug.

Materials and Methods

Materials. Drugs (39) were selected according to their accessibility and varied molecular structures. Drug characteristics, suppliers, and purities are listed in Table 1.

Crystallization Tendency and Thermal Degradation. As no prior information was available on their crystallization tendencies, the studied drugs were analyzed by conventional differential scanning calorimetry (DSC) (Mettler Toledo DSC 823^e, Switzerland) coupled to a refrigerated cooling accessory (Mettler Toledo, METT-FT900 Julabo, Switzerland) and an autosampler (Mettler Toledo, TS0801RO, Sample Robot, Switzerland) to determine the crystallization tendency of melt-quenched samples. The samples were heated from room temperature to 15 °C above the melting point, and held for 5 min. The samples were then cooled to −50 °C and held for 15 min. Finally, samples were heated again to 20 °C above the melting point. A constant heating and cooling rate of 10 °C/min was used. Temperature and heat flow were

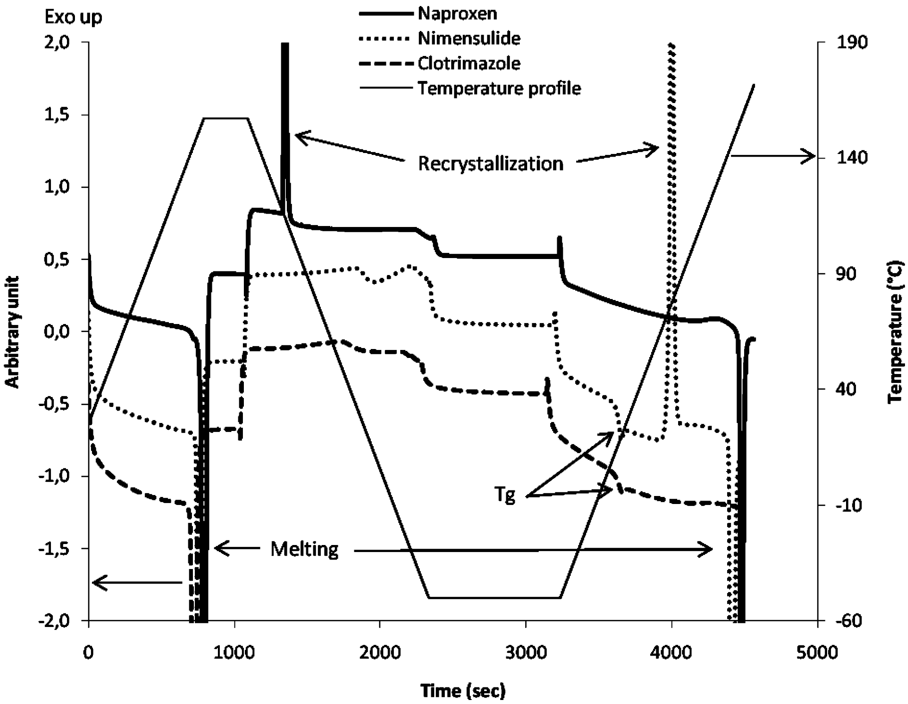

Figure 2. Schematic illustration for categorizing pure compounds, according to their crystallization tendencies. Naproxen represents a typical highly crystallizing compound, nimesulide represents a moderately crystallizing compound, and clotrimazole represents a noncrystallizing compound.

calibrated by using indium, lead, zinc and highly purified water as reference standards. The DSC cell was purged with nitrogen at 50 mL/min. The samples were weighed with an analytical balance (Sartorius SE2, Sartorius AG, Germany), and typical weights of the samples were a few milligrams, depending on the compound's density. The samples were analyzed in sealed 40 μL aluminum pans (Mettler Toledo, Switzerland) with a pierced lid. Three parallel samples of each compound were analyzed. The results were analyzed with STAR^e software (Mettler Toledo, Switzerland). Based on the crystallization of samples during the above-described heat−cool treatment, drug compounds were roughly classified into three categories: *highly crystallizing compounds* crystallized during the cooling segment, *moderately crystallizing compounds* crystallized during the second heating segment, and *noncrystallizing compounds* that did not crystallize at all during the cooling−heating treatment (Figure 2). As the goal was to produce stable amorphous structures from rapidly crystallizing compounds, the compounds having a high crystallizing tendency were considered as drug compounds and the others as stabilizing excipients.

The thermal degradation properties of each drug were studied with a Nicolet Nexus 870 FTIR spectrometer (Thermo Electron Corp, Waltham, MA) equipped with an attenuated total reflectance (ATR) accessory (Smart Endurance, single-reflection ATR diamond composite crystal) and DSC (Mettler Toledo DSC 823^e, Switzerland). The FTIR drug samples were prepared on the lids (PerkingElmer B014-3004) of DSC pans and heated to 5 °C above the melting point with a hot-stage (Linkam Scientific Instruments Ltd., U.K.) light polarizing microscope (Nikon LV100D, Japan) and held there for at least 30 s, and melting was both monitored and verified by polarized light microscopy. Every FTIR spectrum was compiled from 64 scans at a resolution of 4 cm^{-1}. The melting points were determined as onset values and analyzed from the DSC measurements described above. The indicators of thermal degradation were obtained from findings of equality in the FTIR spectrum between heat-treated and untreated samples, and possible melting point depressions from the DSC scans.

Calculation of the Flory−Huggins Interaction Parameter and Phase Diagram. All molecules were geometrically optimized by using the Smart algorithm with ultra fine quality (Materials Studio 4.2, Forcite toolbox, Accelrys Inc. San Diego, CA). COMPASS force-field with force-field assigned charges were used. Summation methods were atom-based for both electrostatic and van der Waals interactions, with no truncation cutoff limits. Interaction parameters were calculated as described in the background section, using the Materials Studio 4.4 Blend toolbox (Accelrys Inc. San Diego, CA). In the interaction parameter calculations, the same force-field settings were used as in geometry optimization. The interaction parameters of 1122 compound pairs were calculated in this research (Table 2 in the Supporting Information).

Validity of Predicted Miscibilities. The validity of the predicted miscibility calculations was studied by using light polarizing microscopy (Nikon LV100D, Japan) equipped with a hot stage (Linkam Scientific Instruments Ltd., U.K.). At this point the selected binary mixtures were roughly divided into two categories: the miscible pairs with negative interaction parameters and those immiscible pairs having a large positive interaction parameter. Additionally, the pairs were subdivided into two groups; in the first group, both the drug substance and excipient had a high crystallizing tendency, and in the second group the drug substance was defined as a highly crystallizing compound but the excipient was either a moderately crystallizing compound or amorphous. It is worth mentioning that all molecules studied in this research were actual drugs, however in the method evaluation some of these molecules were considered as drugs and others as excipients.

To achieve a molecular level mix that has equal mole fractions of molten drug and excipient, both substances were ground gently in mortar and pestle to form a physical mixture and to reduce the particle size. A reduced particle size would be expected to increase the dissolution rate of one miscible compound into the second and hence to promote the occurrence of spontaneous mixing. In a miscible solid binary mix, one compound will normally have a lower melting point than the other. Thus, the lower melting point substance can be considered to act as a solvent while the other can be viewed as the substance to be dissolved. As the miscible compound dissolves in the other substance, the possibility of thermal degradation is reduced because a molten binary mix is achieved at a lower temperature than either of the individual melting points.

A small amount of physical mix was melted on the microscopic hot stage at a heating rate of 30 °C/min. When the powdered mixture was completely melted or dissolved, the object glass was placed on top of it and the sample was allowed to cool at the same rate to room temperature. The crystallization or amorphous phase was then monitored during the cooling phase.

Results and Discussion

Pure Component Crystallization Tendency and Thermal Stability. According to the DSC experiments, the studied drugs were divided into three categories: highly crystallizing compounds, moderately crystallizing compounds and compounds which did not crystallize during heat treatment and remained amorphous (Figure 2). Additionally, melting points and crystallization temperatures were determined as onset values and glass transitions as midpoint values (Table 1). From the 39 studied compounds, 19 belonged to the highly crystallizing category, 6 were moderately crystallizing compounds, and 9 were amorphous in structure. Five compounds degraded simultaneously at the melting temperature, and thus were excluded from further studies. Further FTIR and DSC measurements showed that one more compound (sulfamethoxazole) degraded during heating, and was subsequently excluded from the study. No thermal degradation of the other compounds was apparent in the FTIR spectra or DSC scans (results not shown).

Computational Determination of the Flory−Huggins Interaction Parameter. Interaction parameter calculations started from the determination of the binding energy between the two components and the coordination number (eq 6). The binding energy is a measure of the energy of the interaction between two components, and the coordination number is the maximum number of screen molecules packed around a single base molecule within a set volume (normally the van der Waals surface was used to describe the shape of each molecule) (Figure 3).

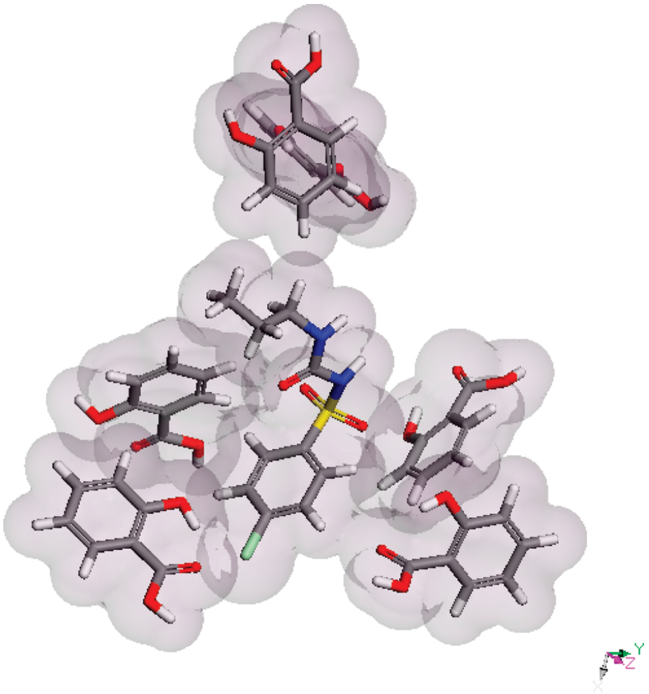

Figure 3. Six most favorable binding configurations between chlorpropamide and salicylic acid (shaded area represents the van der Waals surface).

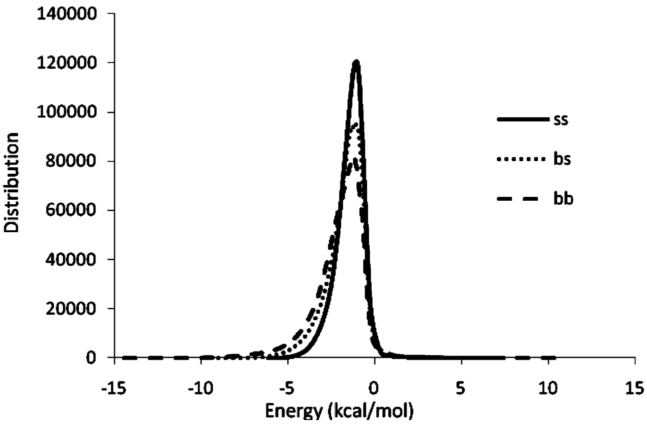

Figure 4. Distribution of binding energies between chlorpropamide and chlorpropamide (bb), chlorpropamide and salicylic acid (bs), and salicylic acid and salicylic acid (ss).

During the binding energy distribution and coordination number calculations, the base molecule was kept fixed and the screen molecule was randomly translated and rotated (via Monte Carlo simulation) in order to obtain a large number pair configurations with a wide range of binding energies and energy distributions (Figure 4).

From the binding energies and coordination numbers, the energy of mixing can be calculated from eq 6. Examples of the mixing energies of chlorpropamide with salicylic acid, and of salicylamide and thiosalicylic acid as functions tem-

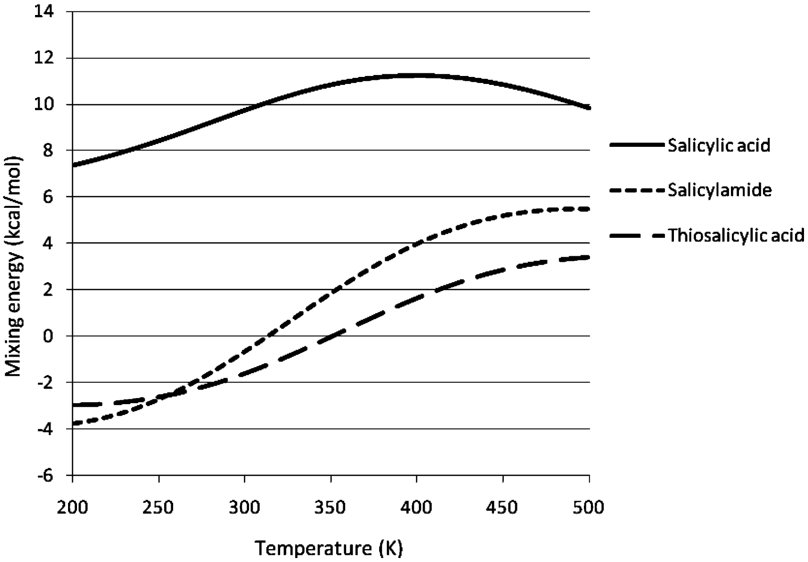

Figure 5. The energy of mixing chlorpropamide with salicylic acid, salicylamide and thiosalicylic acid.

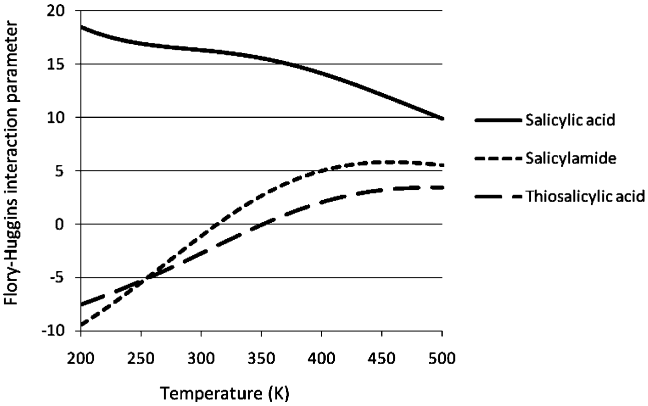

Figure 6. The Flory−Huggins interaction parameter between mixes of chlorpropamide−salicylic acid, chlorpropamide−salicylamide, and chlorpropamide−thiosalicylic acid.

perature are shown in Figure 5, where it can be seen that the mixing energy of chlorpropamide and salicylic acid is positive (endothermic) between 200 and 500 K. In contract, the mixing energies of chlorpropamide−salicylamide and chlorpropamide−thiosalicylic acid are negative (exothermic) up to approximately 320 and 350 K, respectively, after which point it changes to positive. In other words, as spontaneous mixing requires a negative free energy change and the entropy of mixing increases (positively), the enthalpy of mixing has to be negative so that mixing can occur. Based on these results, a chlorpropamide−salicylic acid mix is immiscible over a temperature range of 200−500 K, while mixes of chlorpropamide−salicylamide and chlorpropamide−thiosalicylic acid are miscible up to 320 and 350 K, respectively. At higher temperatures, these components start to phase separate (Figure 5).

From the mixing energy, the temperature dependent Flory−Huggins interaction parameter is obtained by simply dividing the mixing energy by *RT* (eq 5, Figure 6). Once again, the interaction parameter indicated that the chlorpropamide−salicylic acid binary mixture has a large positive value, indicating immiscible behavior. In the cases of chlorpropamide−salicylamide and chlorpropamide−thiosalicylic acid mixes, the interaction parameter values changed from negative to positive at approximately 320 and 350 K, respectively. In other words, mixtures are miscible below these temperatures, whereas phase separation can occur above these threshold temperatures.

When the Flory−Huggins interaction parameter has been solved, the free energy change of mixing, the phase diagram (as a function of composition) and temperature can be determined according to eq 2 (Figures 7 and 8). Figure 7 shows how the free energy of mixing changes as a function of both composition and temperature. The phase diagram gathers all of the obtained data into one figure (Figure 8). Lines represent the presence of coexisting stable and unstable systems. Inside of the curves, a binary mixture is unstable and will phase separate. In contrast, outside of the curves, binary mixtures are thermodynamically miscible and stable.

In Vitro Validation of Miscibility and Phase Stability. In order to achieve rapid screening for the validity of computationally predicted miscibility and phase stability, physical mixtures of highly crystallizing compounds were prepared, melt quenched, and consequently monitored for their crystallization characteristics with hot-stage and polarized light microscopy. Whenever results of a binary mixture are outside the phase diagram curve during the heat treatment, the component possessing the lower melting point melts first and the higher melting point component then dissolves into the first melted component. After all of the crystalline particles dissolved or melted, the temperature was lowered and the appearance of crystallization or solid amorphous phase was monitored.

A total of 26 binary mixtures were randomly selected from the highly crystallizing component category and tested in vitro. At first, pairs were divided roughly into two categories, miscible and immiscible, according to their interaction

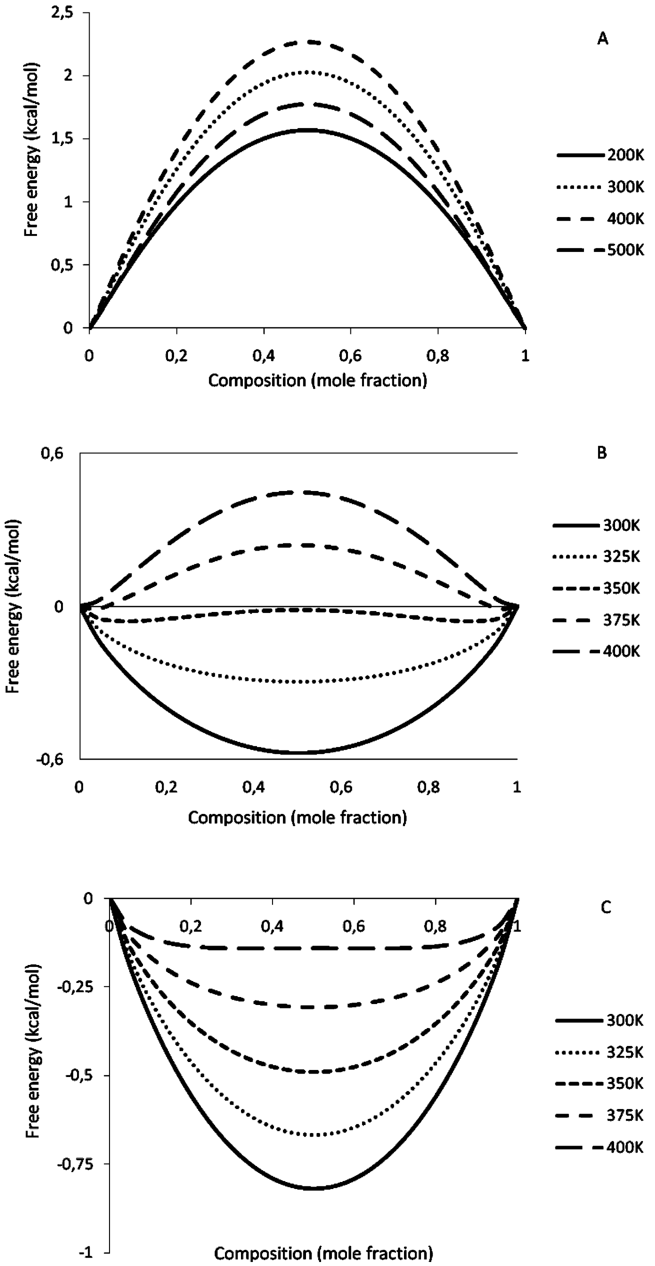

Figure 7. Free energy changes for mixtures of chlorpropamide−salicylic acid (A), chlorpropamide−salicylamide (B), and chlorpropamide−thiosalicylic acid (C).

parameters at 298 K (Table 2 in Supporting Information). However, one must also take into account the temperature dependency of the interaction parameter, which has been estimated from the phase diagram. Hence, miscibility was determined from the phase diagrams (i.e., the temperature range, from room temperature to melting point) and the corresponding Flory−Huggins interaction parameters. Based on this analysis, 15 pairs were predicted to be immiscible and 11 were predicted to be miscible. Through empirical testing, all 15 immiscible binary mixtures crystallized during cooling, as predicted by the phase diagram and interaction parameter. With respect to the miscible binary mixtures, 8 (73%) remained amorphous. The three miscible pairs did crystallize, although according to all calculations (i.e., energy of mixing, interaction parameter, free energy change of mixing and phase diagram) they were predicted to provide an amorphous mixture. However, the interaction parameters were only slightly negative (−0.33, −0.42, −0.45) at the temperature where both compounds were molten. Thus, those binary mixtures lie just within the miscible−immiscible coexistence line. Another possible reason for unexpected crystallization could be inadequate mixing. If the components were not fully mixed to the molecular level, there might be two separate phases coexisting and naturally the phase separation and further crystallization would occur more easily. A third possible explanation for the unexpected crystallization, and more specifically for the partial crystallization, could be the presence of impurities that induce nucleation and crystal growth.

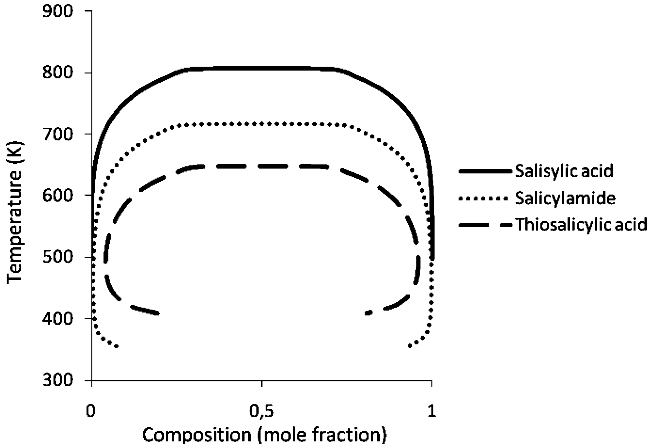

Figure 8. Phase diagrams of chlorpropamide−salicylic acid, chlorpropamide−salicylamide, and chlorpropamide−thiosalicylic acid mixtures.

Next, 20 miscible and immiscible pairs were randomly selected from the moderately crystallizing and amorphous compound categories and studied in vitro. In this case, all binary mixtures remained in the amorphous phase. Thus, the amorphous component in a binary mixture stabilized the system and prevented crystallization, even though the components were immiscible. In these cases, stabilization can be considered to occur similarly as in drug−polymer systems; i.e., an amorphous excipient provides an amorphous mixture.

Although the Flory−Huggins theory seems to predict the miscibility of small molecule binary mixtures relatively well, a clear distinction between miscible and immiscible is not always so apparent (Figure 9). However, when the Flory−Huggins interaction parameters are far from the miscible−immiscible boundary, the interpretation is explicit and the accuracy of predicted miscibilities is very high.

Conclusions

The Flory−Huggins interaction parameter was found to be a relatively good predictor for thermodynamic miscibility and the phase stability of amorphous small molecule binary mixtures. In the analysis of compound pairs, where two

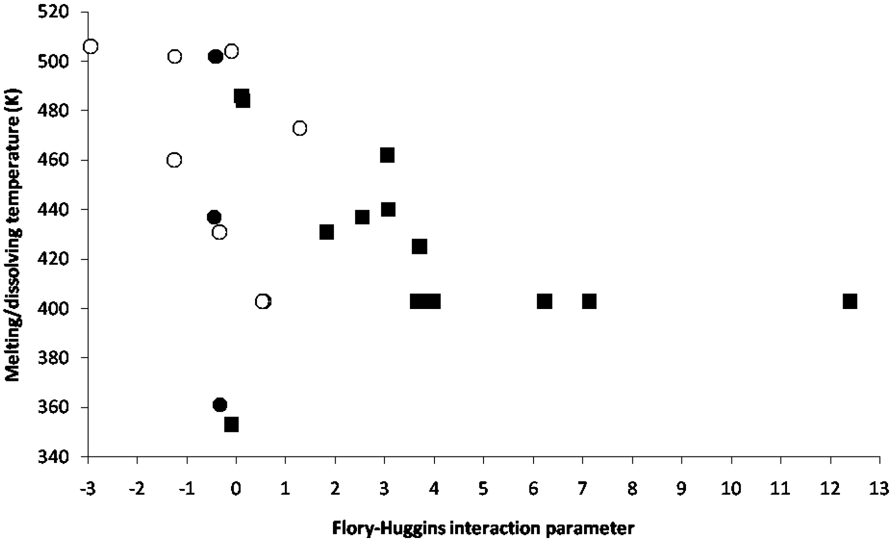

Figure 9. Interaction parameters of in vitro studied pairs for melting temperature. Circles (○ ●) were predicted to be miscible and squares (■ □) immiscible. Open legend indicates amorphous (○ □) and closed legend crystallized binary mixture (● ■).

compounds possessed a high crystallizing tendency, the Flory−Huggins interaction parameter predicted either miscibility or immiscibility leading to amorphous states or crystalline phases, respectively, with 88% confidence. All of the analyzed immiscible pairs crystallized during the experiment, as predicted by the phase diagram. However, 27% of the miscible pairs crystallized despite exhibiting a favorable interaction parameter. This inaccuracy of the miscibility prediction could result from a marginal difference between the calculated and critical interaction parameter;, i.e., the composition of crystallized binary mixtures may have been too close to the thermodynamic line that separates miscible and immiscible mixtures. In addition, the effect of inadequate mixing and the presence of impurities cannot be excluded. All (20) analyzed miscible and immiscible pairs selected from the moderately crystallizing and amorphous compound categories remained in the amorphous phase, which was not unexpected, because a similar method that used polymers as excipients had been widely investigated and used to stabilize amorphous drug compounds. The advantage of calculating interaction parameters to predict binary mixture miscibility is that it provides a truly fast and reliable method to search through a large group of excipients for potential stabilizers of amorphous compounds.

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Supporting Information Available: Calculated interaction parameters at 298 K. This material is available free of charge via the Internet at http://pubs.acs.org.

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