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Toward *In Silico* Prediction of Glass-Forming Ability from Molecular Structure Alone: A Screening Tool in Early Drug Development

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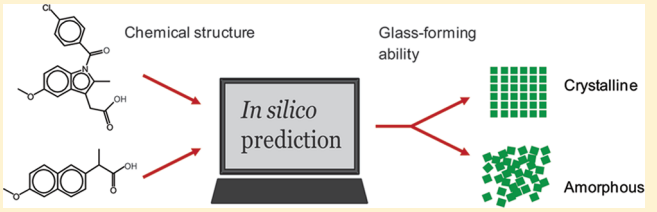
ABSTRACT: We present a novel computational tool which predicts the glass-forming ability of drug compounds solely from their molecular structure. Compounds which show solid-state limited aqueous solubility were selected, and their glass-forming ability was determined upon spray-drying, melt-quenching and mechanical activation. The solids produced were analyzed by differential scanning calorimetry (DSC) and powder X-ray diffraction. Compounds becoming at least partially amorphous on processing were classified as glass-formers, whereas those remaining crystalline regardless of the process method were classified as non-glass-forming compounds. A predictive model of the glass-forming ability, designed to separate between these two classes, was developed through the use of partial least-squares projection to latent structure discriminant analysis (PLS-DA) and calculated molecular descriptors. In total, ten of the 16 compounds were determined experimentally to be good glass-formers and the PLS-DA model correctly sorted 15 of the compounds using four molecular descriptors only. An external test set was predicted with an accuracy of 75%, and, hence, the PLS-DA model developed was shown to be applicable for the identification of compounds that have the potential to be designed as amorphous formulations. The model suggests that larger molecules with a low number of benzene rings, low level of molecular symmetry, branched carbon skeletons and electronegative atoms have the ability to form a glass. To conclude, we have developed a predictive, transparent and interpretable computational model for the identification of drug molecules capable of being glass-formers. The model allows an assessment of amorphization as a formulation strategy in the early drug development process, and can be applied before compound synthesis.



KEYWORDS: glass-forming ability, prediction, in silico models, molecular descriptors, amorphous

1. INTRODUCTION

The major challenges when utilizing the amorphous form of a nonpolymeric organic material are associated with control of its formation and its physical stability, once formed. The choice of components, as well as the method of preparation,[1−4] will have an impact on the degree of disorder and the stability of the processed material. Some organic materials are good glass-formers, i.e. they are intrinsically inclined to become amorphous when solidified by cooling or precipitated from a solution. Conversely, poor glass-formers more easily attain the crystalline state upon solidification. The underlying factors determining a material's glass-forming ability are not well understood, and, in fact, there is an overall deficit of understanding of the physics of the amorphous state.[5,6] Although a theory which links molecular mobility with thermodynamics was suggested by Adam−Gibbs in 1965,[7] a complete theory describing the glass transition has not yet been achieved. Many of the fundamental studies presently reported in the literature have concentrated on describing the dynamics of the liquid-to-glass transformation.[6,8]

Lately, the formation and stability of the amorphous state have been investigated from the perspective of molecular interactions and dynamics.[9−11] There is an increasing awareness that obtaining a better understanding of the glass-formation and amorphous stability is crucial to be able to control the performance of amorphous materials.[5,12] The glass transition temperature (T_g), or, more specifically, the molecular mobility of the amorphous phase, is considered to be an important indicator of physical stability, during both the formation and storage of the amorphous phase since it influences the nucleation and crystal growth rate of the crystalline phase.[13,14] It has become apparent, however, that the T_g alone is not sufficient to describe amorphous stability. Therefore, more recent research has explored the importance of other factors for physical stability, such as localized mobility (β-relaxations),[13,15] as well as considering thermodynamic properties of the amorphous state.[5] In the latter case, it has been suggested that the configurational entropy (or "excess entropy") of a compound is correlated to the amorphous stability.[5,16,17]

The role of hydrogen bonding in the amorphous state for amorphous stability has been evaluated by spectroscopic measurements[18,19] and by correlation to the crystallization enthalpy.[19] Hydrogen bonding has been suggested to increase stability by formation of aggregates that pack poorly and thereby obstruct crystallization.[12] However, the effect of hydrogen bond formation on glass-forming ability is less discussed in literature. Examples of studies can be found in which the chemical structure has been in focus. By introducing specific functional groups Meunier and Lebel managed to convert compounds into good

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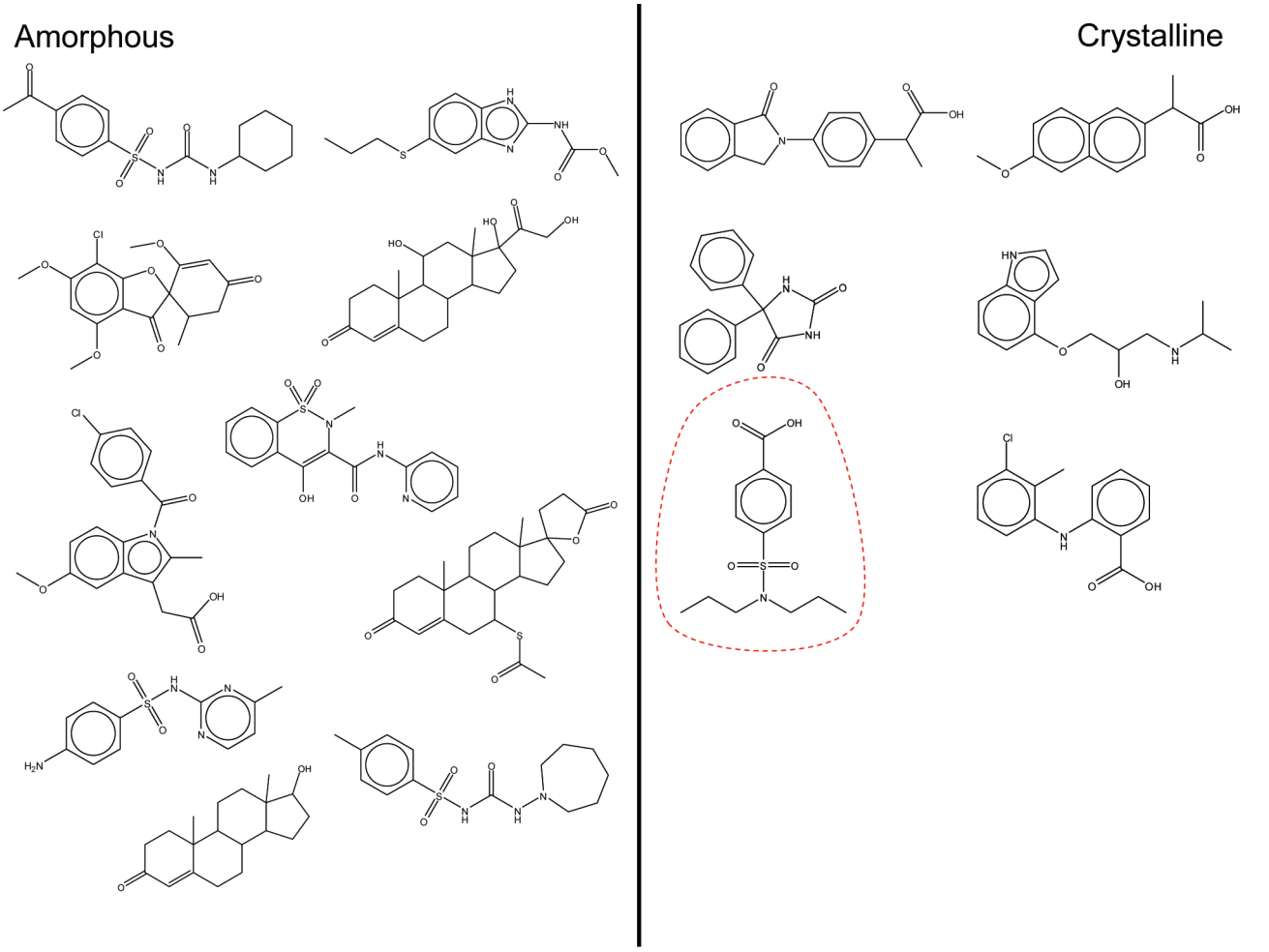

Figure 1. Compounds sorted according to their tendency to form solids in the amorphous or crystalline state when spray-dried. The red dotted circle shows probenecid, which was the compound that was falsely predicted as being amorphous by the PLS-DA model developed.

glass-formers with high amorphous stability.[20] In another recent study, the glass-forming ability has been shown to be dependent on the location and symmetry of the hydrogen bonding groups in butanediols.[21]

Apparently, there are different opinions on the relative importance of the underlying physical factors that govern the amorphous stability during formation and on storage. The inconsistency of the conclusions drawn from the various studies reported could be attributable to the approach generally adopted by researchers in the field: correlations between one isolated factor and stability are usually sought for while other factors are kept constant. In addition to this, generally only a few compounds are included in the studies and, most often, the compounds studied are biased toward good glass-formers. Not until recently have publications appeared that attempt to move away from the "case study" approach: By including 12 compounds, Greaser et al. showed that configurational entropy displayed the best correlation with amorphous stability out of the various thermodynamic and kinetic parameters investigated (R^2 of 0.69).[17] In a study by Lin et al., it was found that the ability to form a disordered material by mechanical activation of 23 small organic molecules could be reasonably predicted by a statistical class model based on the T_g and molecular volume of the compounds.[22] Baird et al. recently made a classification of the glass-forming ability and glass stability of 51 organic molecules upon cooling from the liquid state.[23] The data was used to generate a PCA model that predicted glass-forming ability from the melting temperature, entropy, enthalpy and free energy of fusion, as well as the molecular properties "number of rotational bonds" and molecular weight. Unfortunately, the use of experimentally determined parameters, such as T_g, enthalpy of fusion and melting temperature, as input parameters makes these models inapplicable for early predictions when compounds only are available in limited amounts. Moreover, the models obtained have not yet been challenged with separate test sets, and, therefore, it has not been possible to evaluate their general applicability.

One way to gain control of the amorphous state would be through the development of computational models for the prediction of amorphous state properties from the chemical structure of compounds. Indeed, a molecular understanding of the material properties of drugs would allow early predictions to be made of which manufacturing processes and formulation strategies to apply, for instance, when amorphization of poorly soluble compounds could be applicable. If such predictive tools were to be used in concert with tools for the identification of pharmacologically active leads, rational chemical modifications intended to increase the drug developability could be included in the lead optimization process.

In this work, we studied the ability of pure drugs to form an amorphous state in settings comparable to standard production conditions. A series of 16 compounds were included, selected to be model compounds for drugs displaying solid-state limited aqueous solubility which are likely to gain in apparent solubility by the transformation to the amorphous state, and these were experimentally determined to be either (i) able or (ii) not able to form the glassy state by three different methods applied. The experimental results were used together with rapidly calculated molecular descriptors to develop a class model discriminating between these two groups. The model obtained was accurate, transparent and generally applicable, as proven by challenging it with an external test set. By adopting this approach, it was possible to get a better understanding of a material property—the glass-forming ability—from a molecular structure perspective.

2. METHODS

2.1. Materials. In this study, focus was set on compounds displaying solid-state limited aqueous solubility since amorphization of such compounds is expected to result in significantly increased apparent solubility and dissolution rates. Hence, compounds with high melting points and low lipophilicity, as described by the partition coefficient between octanol and water (log P_{oct}), were prioritized.[24,25] However, to enable the experimental techniques applied to be validated, and to expand the chemical space investigated and thereby make it possible to identify the limits of the glass-forming ability, a few exceptions to these selection criteria were made. For instance, indomethacin, which is a well-known good glass-former,[26] has a relatively low melting point of 162 °C (Table 1) and high log P_{oct} of 4.2 but was included to validate the experimental protocols applied. The final data set (see Table 1) displayed melting points of 152−295 °C (with a median of 201 °C) and had a log P_{oct} of 1.1−4.2 (with a median of 2.6). For chemical structure of the compounds see Figure 1. Finally, to analyze whether the selected 16 compounds were representative of compounds with solid-state limited solubility, their position in the oral drug space was investigated. We have previously established a principal component analysis (PCA) of the chemical diversity of the oral drug space based on the registered oral drugs in Sweden (n = 501; see e.g. Bergström et al.[27]). In Figure 2 this chemical space is shown with compounds showing solid-state limited[25] and solvation limited[27] solubility. When plotting our compounds in the same plot, they clearly cluster with the solid-state limited compounds rather than the solvation limited compounds, and it was therefore concluded that the compound selection was appropriate.

All of the compounds studied were used in their free form, i.e. no salts were included in the analysis and the crystallinity of the starting material was verified by differential scanning calorimetry (DSC, see below).

Acetohexamide was purchased from Fluka, and all the other drugs were obtained from Sigma-Aldrich Chemie GmbH, Germany. The specified purity of all drugs used was >98%, except for griseofulvin (>96%). Ethanol (Alita Corporation, Finland) and acetone (VWR International S.A.S., France) were used as solvents in the spray-drying feed solution.

2.2. Production and Identification of the Amorphous form. *2.2.1. Spray-Drying.* The susceptibility of the compounds to be transformed to the amorphous state was primarily investigated by spray-drying them using a Büchi B-290-mini spray dryer with an inert loop (Büchi Laboratoriums, Switzerland). To allow a higher throughput of the spray-drying technique, we decided to use an optimized protocol applicable to druglike molecules showing solid-state limited aqueous solubility. The parameter setting used in the spray-drying process was obtained through a fractional factorial experimental study design using MODDE 7 (Umetrics, Sweden) based on minimum and maximum values for the following parameters: inlet temperature, spray gas flow, drying air flow, feed pump rate, feed solution concentration and feed solvent composition using griseofulvin and naproxen as model compounds displaying solid-state limited solubility (see Table 2). The optimized method was validated with indomethacin, to confirm that it was able to produce the amorphous form of other known glass-formers than griseofulvin.

Table 1. Physicochemical Characteristics of the Compounds Studied

compound	T_m (°C)[a]	C log P[b]	MW (g/mol)
acetohexamide	188	2.1	324.4
albendazole	208	3.1	265.3
griseofulvin	220	2.2	352.8
hydrocortisone	217	1.2	362.4
indomethacin	162	4.2	357.8
indoprofen	213	2.7	281.3
naproxen	152	2.8	230.3
phenytoin	295	2.1	252.3
pindolol	171	1.9	248.3
piroxicam	198	1.9	331.4
probenecid	194	2.8	285.4
spironolactone	203[c]	3.6	416.6
sulfamerazine	236	1.1	264.3
testosterone	155	3.3	288.4
tolazamide	170	2.5	311.4
tolfenamic acid	207	4.1	261.1

[a] The melting point as reported in *The Merck Index*.[32] [b] Calculated lipophilicity (C log P) by downloading three-dimensional molecular structures from PubChem (http://pubchem.ncbi.nlm.nih.gov/) and use of these as input for molecular descriptor calculation with DragonX version 3.0 (Talete, Milan, Italy). [c] Spironolactone displays a heating rate dependent melting temperature. Hence, the reported value here is the experimentally determined onset of melting from the DSC experiments described in Methods.

Differential scanning calorimetry (DSC6200, Seiko, Japan) was used to determine whether the spray-drying process had produced the compounds in their amorphous form. About 2 mg of the spray-dried material was weighed into an aluminum pan (TA standard pan) with a perforated lid and analyzed at a heating rate of 10 °C/min from 20 °C to a temperature above the compound's melting point while being purged with nitrogen gas at a flow rate of 80 mL/min. The presence of amorphous phase in the samples was judged from the occurrence of an exothermic crystallization peak in the heat flow signal upon heating, alternatively a complete absence of crystallization and melting peaks.

The spray-dried materials were also analyzed to detect any crystal phase present by recording the X-ray diffraction pattern using a Kratzky camera with a linear position-sensitive wide angle detector (Hecus M. Braun X-ray Systems, Graz, Austria) detecting diffracted Cu Kα radiation obtained by an X-ray generator (Philips, PW 1830/40) in a 2θ interval from 17 to 25° (restricted by the limits of the detector). The temperature was controlled by a Peltier element at 25 °C. Samples resulting in spectra with

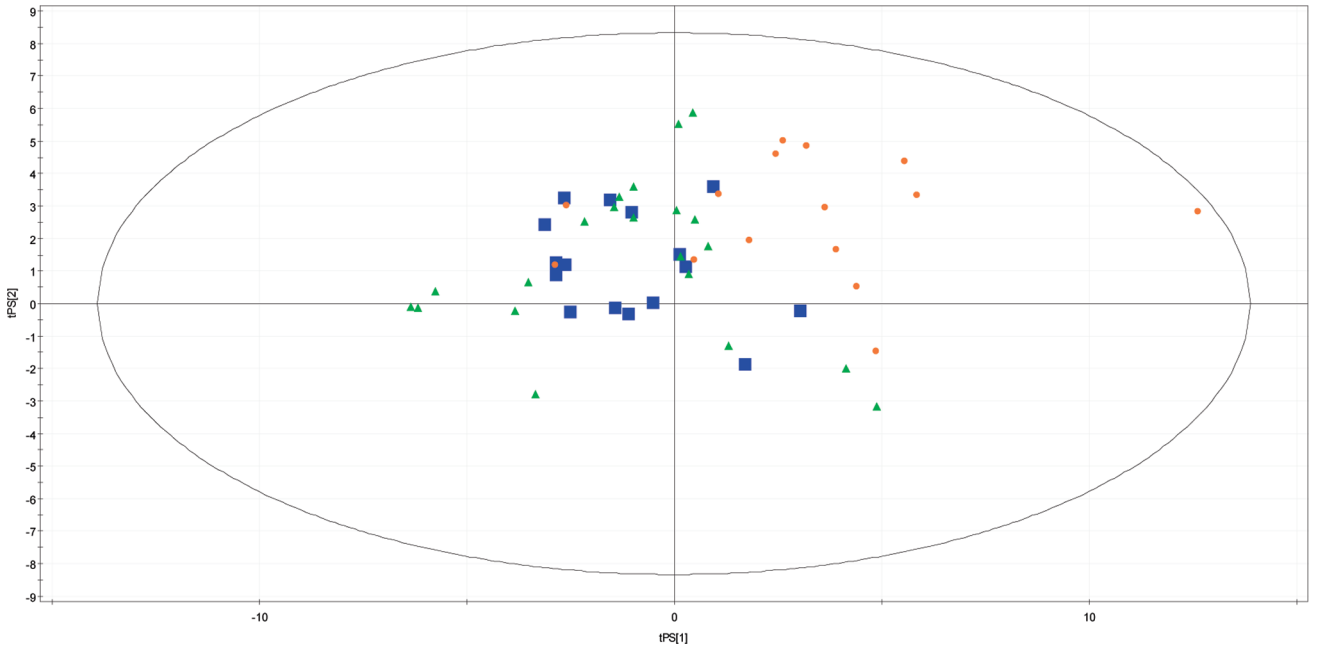

Figure 2. The position of the compounds in the oral drug space. A scatter plot based on principal components 1 and 2 extracted through PCA describing 54% of the chemical diversity of the oral drug space is shown. The investigated data set is shown as blue boxes, and satellite structures of compounds showing solid-state limited compounds[25] and solvation limited[27] and solubility are shown as green triangles and orange circles, respectively. The majority of the investigated compounds were found to be located close to the solid-state limited compounds and hence were regarded as good model compounds for compounds gaining in solubility by amorphization.

Table 2. Spray-Drying Conditions Used in the Design of the Experiment for the Generation of the Spray-Drying Setting, and the Resulting Optimized Values

	value		
parameter	minimum	maximum	optimized
inlet temp (°C)	60	85	50
spray gas flow[a] (L/h)	742	473	608
drying air flow[a] (L/h)	38	31	31
feed pump rate (mL/min)	1.5	11	1.5
feed concn[b] (%)	10	80	80
solvent compos[c] (% ethanol)	10	90	90

[a] Converted from the rotameter value/aspirator rate according to the table provided by the manufacturer. [b] Weight percent of the maximum solubility. [c] Weight percent ethanol in the binary acetone/ethanol mixture.

diffuse background scattering were considered to be amorphous, while samples generating patterns with distinctive sharp peaks were considered to be crystalline. To avoid erroneous conclusions on the solid state due to the limited range of the X-ray detector, it was made sure that the position of major peaks from each compound occurred within the detection limits by comparing their calculated crystalline powder diffraction pattern as generated from the Cambridge Structure Database (Cambridge Crystallographic Data Centre, U.K.).

2.2.2. Melt-Quenching. The ability of the compounds to become amorphous when cooled from the liquid state was investigated by quenching melts of the drugs in the DSC. The experimental setup was the same as for the analysis of spray-dried material, except that about 2 mg of unprocessed substance was weighed into an aluminum pan (TA standard pan and lid) with a perforated lid before being analyzed by performing two heating/cooling cycles, the first for melt-quenching and the second for analysis, in which the samples were heated from room temperature to approximately 10 °C above their melting point at a heating rate of 20 °C/min and immediately cooled at a rate of 40 °C/min. The second cycle was used to judge the presence of amorphous phase in the same way as for the spray-dried compounds.

2.2.3. Mechanical Activation. By mechanical activation a crystalline structure may be transformed into its amorphous form by exposing the solid to mechanical stress.[28,29] In this study, a recently developed method in which the stress is imposed by repeated compressions in a tableting machine[30] was used in parallel with spray-drying and melt-quenching experiments, to study the propensity of the crystalline test materials to transform to, and exist in, an amorphous state. It has been shown that the degree of disorder caused in the solid state increases as a function of applied pressure and the number of compressions.

The fine particulate crystalline form of each compound, as received from the manufacturer, was used as the starting material. In the activation experiments, 30 mg of the test powders was poured into a die and compressed repeatedly by flat-faced cylindrical punches (11.3 mm) mounted in a Korsch (EK 0, Germany) tableting machine. The pressure and number of compressions were systematically altered within a pressure interval ranging from 15 to 300 MPa until a pressure was identified at which the compound became at least partially amorphous. The number of compressions was either 100 or 200. The amorphous content was assessed by DSC in the same way as for the spray-dried compounds, except that a sample weight of 5−10 mg was used. If a sample was judged amorphous after repeated compression, based on the presence of a crystallization peak in heat flow signal, the compound was judged as a glass-former by mechanical activation.

2.3. Computational Modeling. A predictive model of glass-forming ability was developed through the use of partial least-squares projection to latent structure discriminant analysis (PLS-DA) in Simca v.11 (Umetrics, Sweden). The computational model was designed to differentiate between glass-formers, i.e. compounds able to form its amorphous state, and non-glass-formers, i.e. compounds remaining crystalline after processing, from calculated molecular descriptors alone. The descriptors were obtained from DragonX (Talete, Italy), and 245 variables were used as input while the response variables were the two classes "glass-formers" (assigned value 0) and "non-glass-formers" (assigned value 1). The three different methods used for producing the amorphous state had equal weight when sorting the compounds into these two classes. If two out of the three materials produced from each compound were amorphous, the compound was sorted as a "glass-former", whereas if two out of three were crystalline, the compound was sorted as a "non-glass-former". This classification took into account neither how much of the bulk became amorphous upon processing nor whether the amorphous material was stable over time; only the ability to exist in the amorphous state after conventional material processing was modeled.

The data were mean centered and scaled to unit variance. A reduction of the number of variables was done to decrease the complexity of the models, facilitate interpretation and increase the accuracy. First, the bottom 50% of the descriptors, exhibiting the lowest level of importance, was excluded. Second, descriptors duplicating the information contained within other descriptors and therefore residing in the same area of the PLS loading plot were excluded, to leave just four of the original 245 variables in the final model. The aim of this variable selection was to increase the predictivity and robustness of the model by removing information that was not directly related to the response investigated (i.e., noise). The accuracy of the PLS models was judged by how well the two classes of the training set were separated from each other. Once the selection of descriptors had been finalized, the resulting model was validated with a test set of 16 compounds for which the glass-forming ability was reported in the literature.[22]

2.4. Cambridge Structural Database (CSD). The Crystal Structure Database (CSD) from Cambridge Crystallographic Data Centre, U.K., was used to visualize the 3D molecular structure of the compounds investigated to gain an understanding of how the compounds interact with neighboring molecules in the solid state. This information was used, together with the *in silico* model developed, to interpret the outcome of the PLS-DA model and, hence, the glass-forming ability of the compounds.

3. RESULTS

3.1. Preparation of Amorphous Materials. *3.1.1. Spray-Drying.* Naproxen and griseofulvin were used in a multivariate study design to find a spray-drying procedure that was able to produce powders of the compounds with a high degree of amorphous content. The idea was to use the parameter setting obtained from the multivariate optimization as a standardized procedure in the evaluation of the ability of compounds to be amorphized by spray-drying. Naproxen was found to become crystalline independent of process parameters used. The degree of disordering of griseofulvin, on the other hand, was dependent on the parameter setting. The most significant factors affecting the amorphous content were the inlet temperature and the concentration of the feed solution. The optimized setting (Table 2) was applied to spray-dry indomethacin, which resulted in formation of an amorphous powder. The method developed was hence shown to be able to make good glass-formers amorphous, and proven not only to be useful for the production of amorphous griseofulvin. It was thereafter used in the investigations of 13 more drug compounds, of which all turned into fine powders upon spray-drying. For eight of the 16 compounds in total, the product was crystalline as detected by DSC and X-ray diffraction. All other compounds turned out to be either partially or completely amorphous and, hence, were classified as "amorphous after spray-drying", see Table 3. Based on the crystallization and melt enthalpy values, an estimation of the amorphous content of the spray-dried powders was made.

Table 3. A Summary of the Solid State of the Compounds after Processing, and the Overall Classification of Compounds As Glass-Formers or Non-Glass-Formers[a]

compound	after spray-drying	after melt-quenching	after mechanical activation	glass-former?
acetohexamide	●	●	●	yes
albendazole	●	●	●	yes
griseofulvin	●	●	●	yes
hydrocortisone	●	●	●	yes
indomethacin	●	●	●	yes
testosterone	●	●	●	yes
piroxicam	●	●	●	yes
spironolactone	●	●	●	yes
sulfamerazine	○	●[b]	●	yes
tolazamide	○	●	●	yes
phenytoin	○	●[b]	○	no
indoprofen	○	○	○	no
naproxen	○	○	○	no
pindolol	○	○	○	no
probenecid	○	○	○	no
tolfenamic acid	○	○	○	no

[a] (●) Amorphous phase detected; (○) only crystalline phase detected. [b] Indication of decomposition by color change (to dark brown/black) on melting.

According to this estimation hydrocortisone, indomethacin and spironolactone were fully amorphous; griseofulvin and testosterone were approximately 85% amorphous; and acetohexamide, albendazole and piroxicam were about 50, 13 and 7% amorphous, respectively. It should be emphasized though that these values are not precise, but rather should be treated as rough estimations. Further, the X-ray diffraction confirmed the solid state structure of the compounds after spray-drying.

3.1.2. Melt-Quenching. All compounds melted close to the value of the melting point reported in the literature, indicating that all were crystalline and in their most stable crystal form when received. The compounds lacking an exothermic crystallization peak upon subsequent cooling were classified as "amorphous after melt-quenching" (see Table 3). None of these compounds exhibited either a crystallization peak or a melting endotherm upon second heating in the DSC, which indicates that the amorphous material was relatively stable after the melt-quenching.[23] The compounds that crystallized during the cooling stages in the temperature program were classified as "crystalline after melt-quenching".

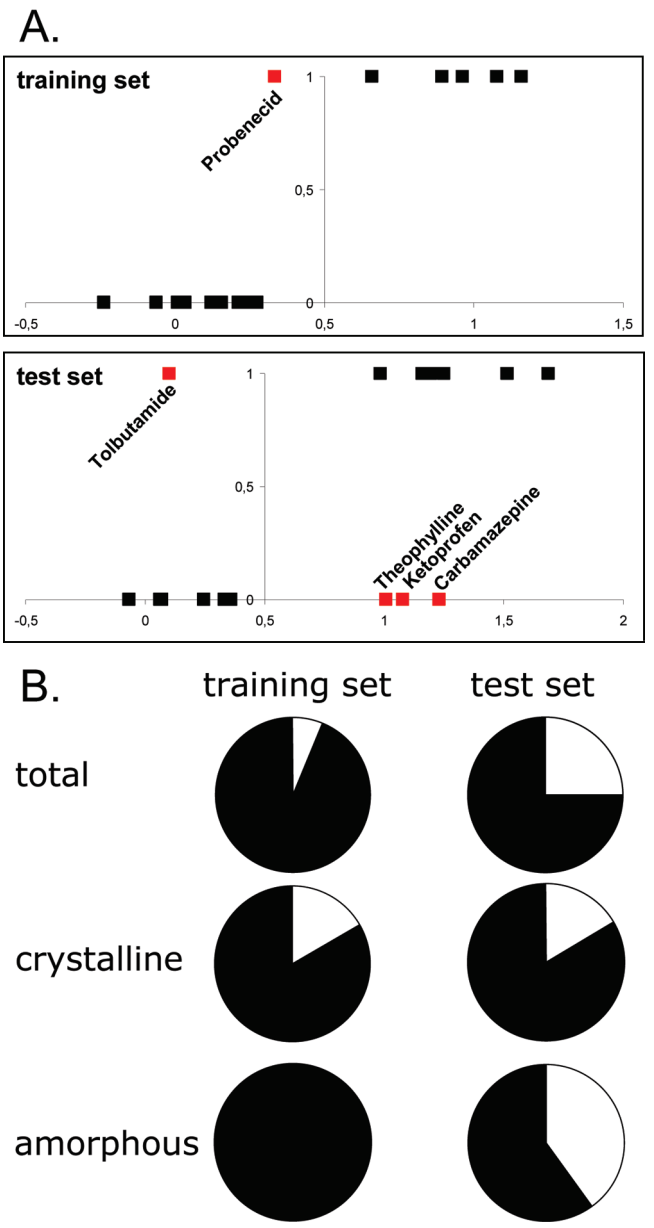

Figure 3. The accuracy of the classification of the training set and the test set used. (A) The compounds were denoted 1 if experimentally determined as crystalline and 0 if amorphous; hence the cutoff between these classes in the prediction was 0.5. The falsely predicted compounds are colored red. (B) Pie charts showing correct and false predictions of glass-forming ability by the developed PLS-DA model. The correct classification is shown in black, false predictions in white.

3.1.3. Mechanical Activation. As was the case after spray-drying and melt-quenching, the amorphization potential after exposure to mechanical stress was judged from DSC thermograms of the mechanically stressed samples. The results of the mechanical activation experiments were consistent with the two other methods of preparation of amorphous materials (see Table 3). In the cases of phenytoin, sulfamerzine and tolazamide, all three of which responded differently to spray-drying and melt-quenching, the latter two were amorphized by mechanical activation, whereas phenyton remained crystalline. Overall, the mechanically activated compounds had a much smaller amorphous content than their spray-dried counterparts as determined by comparing the heat of crystallization in the DSC thermograms for the mechanically activated and spray-dried materials.

3.2. Computational Modeling. A class model with the ability to identify glass-formers from molecular structure was developed by use of PLS-DA. To certify that the model would predict the inherent glass-forming ability and avoid a process dependency, the compounds were assigned their glass-forming ability on the basis of the outcome of the three different experimental processes used. The training set consisted of the 16 compounds of which 10 were classified as glass-formers (see Table 3). The PLS-DA correctly sorted 15 of the 16 compounds with only probenecid, which remained crystalline after all three processes, being misclassified by the computational model as having a glass-forming ability (Figure 3A). To assess the general applicability of the model and to certify that the model was not biased to discriminate the compounds in the training set only, the model was challenged with a test set of 16 compounds taken from the literature. Of these, 12 were correctly sorted by the model.

The descriptors proven to be of importance for the glass-forming ability of the chemical space investigated herein were the number of benzene rings (which was negatively correlated to glass-forming ability), and the branching of the carbon skeleton as described by the XMOD (Modified Randic Connectivity Index), the molecular symmetry as described by VAR (Variation) and the distribution of electronegative atoms in the molecule as described by the Whete (Wiener-type index from electronegativity weighted distance matrix) descriptor, the latter three positively correlated to glass-forming ability. This means that the higher the number of benzene rings, the higher the probability that a compound will remain in the crystalline state on processing, and highly branched, asymmetric molecules that contain electronegative atoms have a higher tendency to form the amorphous state. The descriptors XMOD, VAR and Whete are all three linked to the molecular size in such a way that the numeric value of these descriptors will increase with an increase in the molecular size.

4. DISCUSSION

4.1. Classification of Glass-Forming Ability. The three methods used in this study represent three fundamentally different ways to make compounds amorphous, i.e. from the liquid state, from solution and by the application of mechanical stress. It was, therefore, not surprising that some of the compounds exhibited different responses, and therefore did not receive the same classification by all three production methods. However, the outcome was process-dependent only for three of the compounds, which indicates that the nature of the compound is often more decisive for its amorphization potential than the means of preparation.

In the subsequent modeling of the glass-forming ability, it was useful to have a binary outcome, i.e. compounds being either "a glass-former" or "a non-glass-former". Therefore, the overall classification was made on the basis of the outcome of all three methods (see Table 3), where the process dependent compounds were classified as amorphous if two of the methods resulted in amorphous material (tolazamide and sulfamerazine), and vice versa for the predominantly crystalline outcomes (phenytoin). In the latter case, the melt-quenching method suggested phenytoin to be an amorphous powder. The decomposition of phenytoin at elevated temperatures, like the one used for producing the melted form of the compound, is well-known

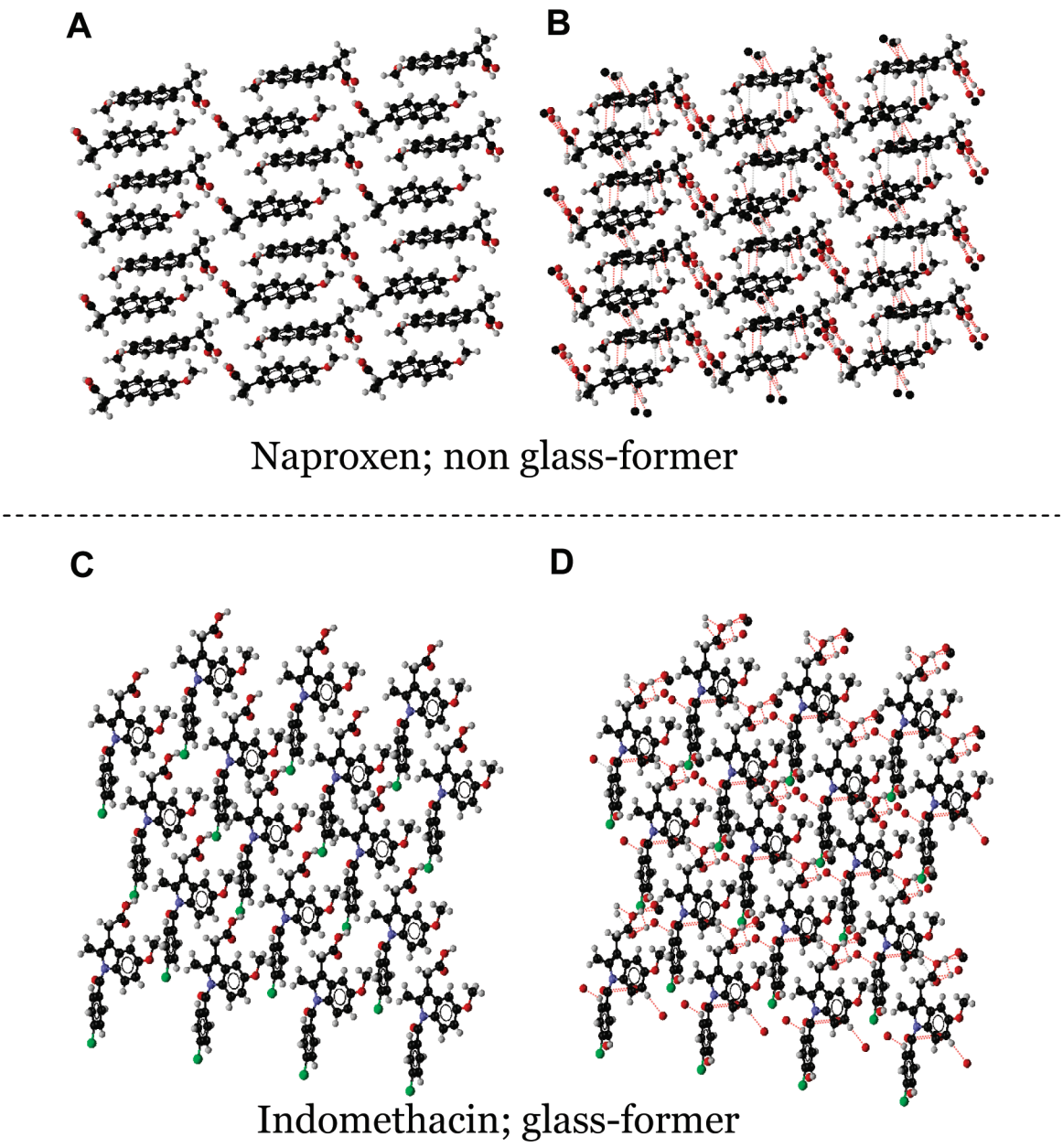

Figure 4. Crystal structures of the non-glass-former Naproxen and the glass-former Indomethacin. (A) and (C) shows respective structure without indicating intermolecular interactions, whereas (B) and (D) shows the structures with van der Waals interactions and intermolecular hydrogen bonds..

(e.g., U.S. Pharmacopeia) and, indeed, the phenytoin observed after melting was dark brown. Our approach to use three different preparation methods substantially reduced the risk of an erroneous classification of the compounds, and thereby making the input matrix for the computational modeling as robust as possible.

4.2. Computational Modeling. The separation between glass-formers and non-glass-formers was excellent for the training set used in the study. To certify that the model had not been overtrained and only applicable to the 16 compounds included in our data set, we challenged the model with a test set taken from the literature. However, it was not an easy task to select the test set, since few papers include a large enough data set based on the same method of preparation. The test set applied was taken from Lin and co-workers and consisted of 16 compounds.[22] The compounds had been processed by cryomilling to produce the amorphous material and had been kept at −50 °C before the analysis of the amorphous content. Some of the compounds in this data set were pharmaceutical excipients and not drugs (e.g., sucrose, lactose, mannitol and trehalose), but were included to obtain as many compounds as possible in the test set. The computational model correctly sorted 75% of the compounds based on the four molecular descriptors (Figure 3B). Most impressively, it had the capacity to separate between the sugars mentioned above, and correctly sorted the glass-forming sugars from the crystalline ones. The compounds that were not correctly predicted in the test set were tolbutamide, theophylline, ketoprofen and carbamazepine, of which the latter three were observed by Lin et al. to form amorphous material, whereas our model predicted them to be crystalline. As previously mentioned, the model presented herein predicts the glass-forming ability at

room temperature. However, ketoprofen has a reported T_g of −4.5 °C and hence the three methods we used, which were designed to measure amorphous content at room temperature, would be likely to produce the crystalline form of this compound. To summarize, we found the novel *in silico* model to be predictive, even for compounds that had not been in the training set and had been amorphized by a method other than those used for generating the data. Although the data set so far studied is small (16 + 16 compounds), the results are very promising with regard to allowing prediction of glass-forming ability of pharmaceutical materials (drugs and excipients) from molecular descriptors solely.

To better understand the molecular structural features that govern the glass-forming ability, a qualitative interpretation was made, based on the four significant descriptors of the PLS-DA model in combination with the structural information obtained from CSD. For instance, the realization that an increased number of benzene rings will lead to a poor glass-forming ability may be related to that fact that such molecules are generally planar and flat and, for this reason, quite easily packed together in a crystal. The strong van der Waals interactions between the benzene rings in the different layers of the crystal should further strengthen the formation of the crystal. This is in agreement with previous models of the crystal energy.[31] The more branched and less symmetric molecules more easily form an amorphous state, as evident from the XMOD and VAR descriptors, respectively. When the 3D crystal structure of these compounds is compared to less branched and more symmetric molecules, the former is seen to have a much more complex interaction pattern, with a large number of possibilities for the formation of intermolecular bonds (Figure 4). For such compounds a more random looking interaction pattern is revealed. In addition to this, the distribution of electronegative atoms seems to be of importance, as indicated by the significance of the Whete descriptor. This may actually reflect the role of hydrogen bonding pattern, which has previously been shown to be of importance for glass-forming ability.[21]

5. CONCLUSIONS

The PLS-DA model presented herein is the first of its kind to predict the glass-forming ability of druglike molecules. Computational tools as the one presented have the potential to be applicable in the early drug development setting for the prediction of compounds that successfully can be transformed to the amorphous state. Such information is valuable when evaluating the possibility to formulate poorly soluble compounds into functional products, and thereby avoiding premature termination of drug candidates due to expected solubility problems. The *in silico* model presented was based on molecular descriptors reflecting the aromaticity, symmetry, distribution of electronegative atoms, branching of carbon skeleton and size, and was predictive for an external test set that had been processed by milling, yet another method than those we used in this study. This shows that a successful transformation of compounds to the amorphous state is strongly reliant on the inherent glass-forming ability of each compound and, to a lesser extent, on amorphization method. However, the general applicability of the model developed needs to be further tested, and additional investigations are needed to allow an increased understanding of the information that each molecular descriptor carries, as well as of the issue of physical stability of the amorphous phase on storage. We are, therefore, currently expanding our data set used for model development to allow future investigations of physical stability of the amorphous state from a molecular perspective.

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