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Señor Director
DIRECCIÓN DE NUEVAS CREACIONES
SUPERINTENDENCIA DE INDUSTRIA Y COMERCIO
Bogotá, D.C.

Re.: Expediente No. 09 149.186
Fase nacional de Solicitud de Patente PCT
a nombre de ACTIMIS PHARMACEUTICALS, INC.
Código 011 - 378 - 444

En relación con la solicitud contenida en el expediente de la referencia y, en particular, para dar respuesta al Concepto Técnico contenido en su último Oficio No. 14175, proferido dentro del mismo, dentro del término conferido, por medio del presente escrito me permito manifestarle:

1. Mi representada me ha dado precisas instrucciones de presentar un nuevo capítulo de reivindicaciones.
2. En consecuencia, acompaño al presente escrito las reivindicaciones 1 a 24, junto con el comprobante de pago de la tasa correspondiente a catorce (14) reivindicaciones adicionales a partir de la número 10, Recibo No. 141.067
3. Con respecto a las modificaciones hechas a las reivindicaciones nos permitimos hacerle notar:
 - a) La reivindicación 1 se modifica acogiendo las observaciones del señor Examinador.
 - b) Se eliminan las reivindicaciones 19, 28, 38 y 47.
 - c) Se elimina el término "aproximadamente" de las reivindicaciones que lo contienen y las reivindicaciones que contienen los términos "solvente", "temperatura predeterminada", "anti-solvente" se eliminan del capítulo de reivindicaciones.
4. Ahora bien, el señor Examinador considera que los documentos WO 2004/096777 D1 y Farmacia Remington D2 afectan el nivel inventivo de las reivindicaciones. Sin embargo, mi representada considera que las reivindicaciones que se acompañan cumplen con el nivel inventivo frente a los documentos citados, por cuanto:

- 4.1 La reivindicación independiente 1 está dirigida a un polimorfo de una sal etilendiamina del ácido {4,6-bis(dimetilamino)-2-(4-(4-(trifluorometil)-benzamido)bencil-pirimidin-5-il} acético (en lo sucesivo, "el Compuesto"), el cual tiene un patrón de difracción en polvo de rayos X con un pico expresado en dos-theta a aproximadamente 5,9°.
- 4.2 En contraposición, D1 está dirigido a pirimidinas y revela el Compuesto solamente como uno de los 147 compuestos específicos que allí se describen. Ver, p.ej., página 8, línea 22 a 18, línea 6 de D1. Sin embargo, D1 no enseña ni sugiere una sal etilendiamínica de ningún compuesto de los allí revelados, mucho menos un polimorfo de una sal etilendiamínica del Compuesto que requiere un patrón de difracción en polvo de rayos X con un pico expresado en dos-theta a aproximadamente 5.9°, como el aquí reivindicado. Las deficiencias de D1 no pueden ser subsanadas por D2, porque D2 solamente ofrece enseñanzas generales de las ciencias farmacéuticas.
- 4.3 D1 y D2 tampoco ofrecen motivación a la persona medianamente versada en la materia técnica correspondiente para llegar a la sal etilendiamina cristalina reivindicada del Compuesto. Sin la enseñanza de la solicitud en estudio, la persona medianamente versada en la materia técnica correspondiente no tendría una expectativa razonable a partir de las enseñanzas de D1 y D2 de que se pudiera haber realizado una sal etilendiamínica cristalina del Compuesto. Por ejemplo, el Compuesto contiene cuatro grupos amino básicos (es decir, 2 grupos dimetilamino y 2 nitrógenos del anillo de pirimidina) y un grupo ácido del ácido carboxílico, y por ello, el Compuesto puede formar una sal zwitteriónica, una sal de adición de base con una base; o una sal de adición de ácido con un ácido. Para formar una sal con una base amina, la base amina tiene que competir con por lo menos uno de los cuatro grupos básicos del Compuesto por su grupo del ácido carboxílico. A causa de esta complicación, la gran mayoría (14 de 18) de los compuestos de amina examinados en la solicitud en estudio no forma sales cristalinas con el Compuesto apropiadas para usos farmacéuticos. Ver la Tabla 1 en las páginas 41 y 42 del PCT '781.
- 4.4 Además, es bien sabido que las técnicas químicas suelen ser impredecibles. Esto es especialmente cierto en el caso de los polimorfos de un compuesto, incluidos los polimorfos de las sales. Ver Peterson et al.,

"Expanding the Scope of Crystal Form Evaluation in Pharmaceutical Science" *J. Pharm. Pharmaceut. Sci* 2006, 9, 317-326 ("no hay manera de predecir la extensión práctica del polimorfismo de ningún compuesto dado"); Bernstein, "Crystal Structure Prediction and Polymorphism" *ACA Transactions* 2004, 39, 14-23 ("todavía queda mucho por conocer y hacer antes de poder afirmar con algún grado de confianza que la capacidad de predecir una estructura cristalina a partir de una fórmula estructural sola, mucho menos de sus formas polimórficas") Por consiguiente, habría sido imposible predecir si una sal etilendiamina del Compuesto sería cristalina o un sólido amorfo.

4.5 Así pues, sin la enseñanza de la presente solicitud, la persona medianamente versada en la materia técnica correspondiente no tendría una expectativa razonable a partir de las enseñanzas de D1 y/o D2 de que la sal etilendiamínica cristalina del Compuesto aquí reivindicado pudiera haber existido. Por ello, la reivindicación independiente 1 y sus reivindicaciones dependientes tienen nivel inventivo por sobre D1 y D2, solas o en combinación.

4.6 La reivindicación independiente 6 está dirigida a un polimorfo de una sal piperacínica del Compuesto, la cual tiene un patrón de difracción en polvo de rayos X con uno o más picos expresados en dos-theta a aproximadamente 10,7, 15,9, 22,3, o 24,0°. Sin embargo, D1 no enseña ni sugiere una sal piperacínica de ningún compuesto de los allí revelados, mucho menos un polimorfo de una sal piperacínica del Compuesto que requiere un patrón de difracción en polvo de rayos X con uno o más picos expresados en dos-theta a aproximadamente 10,7, 15,9, 22,3, o 24,0°. Por consiguiente, por las mismas razones que aquí se expresaron para la reivindicación 1, la reivindicación independiente 6 y sus reivindicaciones dependientes tienen nivel inventivo por sobre D1 y D2, solas o en combinación.

4.7 La reivindicación independiente 12 está dirigida a un polimorfo de una sal benzatínica del Compuesto, la cual tiene un patrón de difracción en polvo de rayos X con uno o más picos expresado en dos-theta a aproximadamente 8,0, 11,5, 16,0, 17,5, o 23,4°. Sin embargo, D1 no enseña ni sugiere una sal benzatínica de ningún compuesto de los allí revelados, mucho menos un polimorfo de una sal benzatínica del Compuesto que requiere un patrón de difracción en polvo de rayos X con uno o más picos expresado en dos-theta a aproximadamente 8,0, 11,5, 16,0, 17,5, o 23,4°.

4

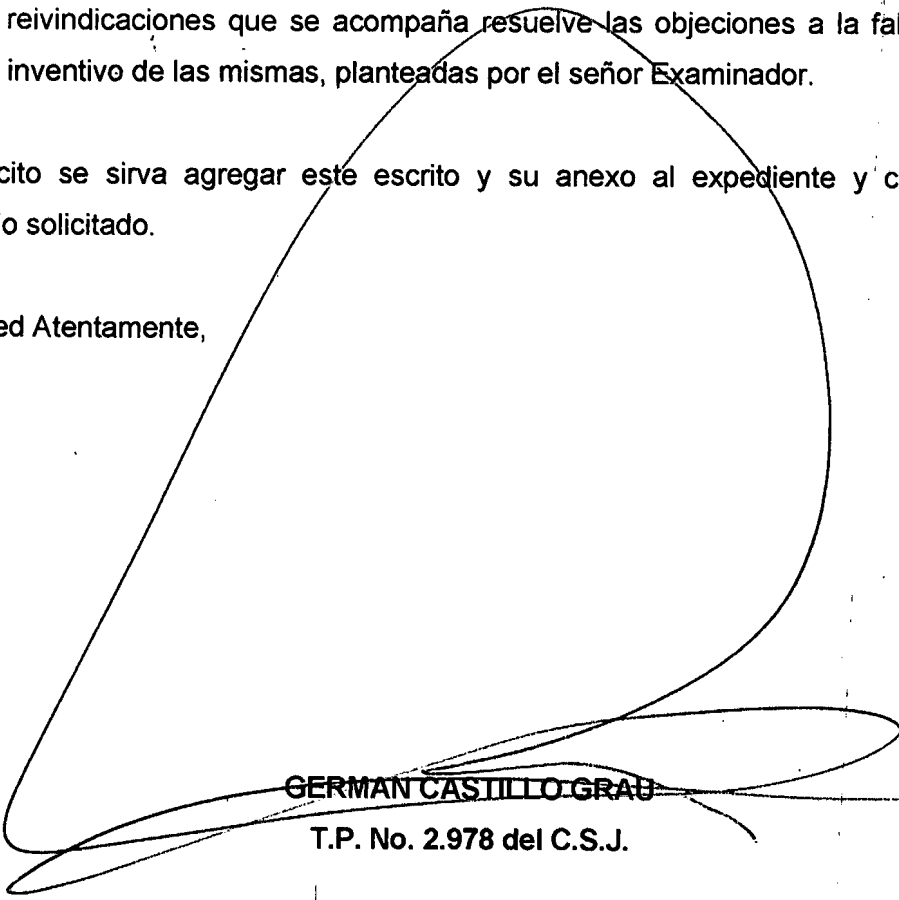
Por consiguiente, por las mismas razones que aquí se expresaron para la reivindicación 1, la reivindicación independiente 12 y sus reivindicaciones dependientes tienen nivel inventivo por sobre D1 y D2, solas o en combinación.

4.8 La reivindicación independiente 17 está dirigida a un polimorfo de una sal de colina del Compuesto, la cual tiene un patrón de difracción en polvo de rayos X con uno o más picos expresados en dos-theta a aproximadamente a 6,5, 19,6, 20,0, 21,9, o 26,1°. Sin embargo, D1 no enseña ni sugiere una sal de colina de ningún compuesto de los allí revelados, mucho menos un polimorfo de una sal de colina del Compuesto que requiere un patrón de difracción en polvo de rayos X con uno o más picos expresados en dos-theta a aproximadamente 6,5, 19,6, 20,0, 21,9, o 26,1°. Por consiguiente, por las mismas razones que aquí se expresaron para la reivindicación 1, la reivindicación independiente 17 y sus reivindicaciones dependientes tienen nivel inventivo por sobre D1 y D2, solas o en combinación.

5 Consideramos que con las correcciones efectuadas, el capítulo de reivindicaciones que se acompaña resuelve las objeciones a la falta de nivel inventivo de las mismas, planteadas por el señor Examinador.

Le solicito se sirva agregar este escrito y su anexo al expediente y conceder el privilegio solicitado.

De Usted Atentamente,


GERMAN CASTILLO GRAU

T.P. No. 2.978 del C.S.J.

CRYSTAL STRUCTURE PREDICTION AND POLYMORPHISM

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1. ABSTRACT

to predict: to foretell, prophesy, announce beforehand
Oxford English Dictionary

Since the early days of structure determination by diffraction methods the prediction of crystal structures of small to medium-sized organic molecules from a structural formula has been a goal and a constantly recurring theme of crystallographers. The challenge is even greater when considering the additional factor of the possible existence of polymorphic forms of the material, since true predictive capability requires accounting for all crystal forms and their relative stability. While considerable progress has been made and some practitioners have even claimed that the goal has been achieved, a great deal still needs to be learned and done before one can state with any degree of confidence the ability to predict a crystal structure from a structural formula alone, much less its polymorphic forms. An even greater challenge is the prediction of solvates, including hydrates. This contribution attempts to summarize some of the current thinking on the subject.

2. INTRODUCTION

At the outset to this contribution, I want to make it clear that I am an enthusiastic believer in the importance of computational approaches to the understanding of interactions within molecular crystals and their potential for aiding in the solution of crystal structures, as well as for surveying the energy landscape for possible, even as yet unknown, structures. I have been following the development of this field for nearly forty years, even with occasional personal forays into it. I am impressed by the progress that has been made, and I enthusiastically support and encourage those who are attempting to develop and refine the field in general and its applications to polymorphs in particular. The rapidly increasing interest in polymorphism in recent years, from the purely scientific point of view to the commercial and intellectual property aspects has led to increasing (perhaps more correctly intense) interest in our ability to predict *ab initio* the existence of multiple crystal forms for a particular compound. I do think that some of the hyperbole and optimism of the past decade concerning our ability to predict even the possible existence of polymorphs was misplaced, and this paper is an attempt to present my perspective on the current situation. Although I have attempted to be objective, this minireview is the view from one person's perspective, which must per force carry some bias.

As with many aspects of chemistry, much discussion and debate surround our definition of terms, and the associated frame of reference. In this case, what do we mean by the term "to predict"? The Longman's Dictionary has a slightly different definition from that given by the OED above: "to see or describe in advance as a result of knowledge, experience, thought, etc." Using classical physics we can predict the path of a cannon shell or an interplanetary space probe. In chemistry we can often predict the outcome of a reaction or the bonding geometry around an atom. In the current context I view a prediction as the ability to see or describe in advance a crystal structure simply on the basis of its molecular structure. The key phrase in this definition is 'in advance'. For a sound scientific prediction we don't have to do the experiment to know what the result will be. It may be technologically challenging or difficult to carry out the experiment, but we expect the known scientific truths to be valid and the accepted scientific laws to be obeyed. By either dictionary definition above the notion of a prediction in scientific terms carries with it some degree of confidence, implied in the Longman's definition "as a result of knowledge, experience, thought, etc." That is, we expect the prediction to be proven.

In the context of the historical development of "predicting" crystal structures of small to medium molecules, 1994 was a particularly notable year, with two nearly simultaneous publications. One was the official announcement by Molecular Simulations Inc. (now Accelrys) in July of the availability of the "Polymorph Predictor" program [1]. The second event was the publication in October of the stimulating paper by Angelo Gavezzotti [3] entitled "Are crystal structures predictable?" to which he gave the emphatic one word opening response "No!" The dichotomy is glaring. How can there be a program for *predicting* multiple crystal structure (i.e. polymorphs) if we can't even predict a single crystal structure? Part of the problem arises, of course, from how each author interprets of the definition of *predict*, and we will attempt to address that aspect of the question as well as making a general assessment of the current situation as seen from the perspective of this viewer.

3. SOME HISTORICAL BACKGROUND

In the current context of automated CCD diffractometers and "black box" programs for the solution, refinement and presentation of crystal structures it may be difficult to imagine (or to recall) the days when the few text books on crystal structure analysis (e.g. 4,5) contained whole chapters on "Trial and Error Methods" for solving crystal structures, more correctly for providing the initial model. In many ways these were essentially means for describing the crystal structure in advance as a result of knowledge experience, thought, etc., i.e. predicting the crystal structure. Much thought and effort went into these methods, since the cost in time and work (often on the order of months) in getting it wrong could be frustrating, if not disastrous. With the advent of powerful desktop or laptop computers armed with efficient software for structure solution (generally by direct methods) and refinement, those chapters have disappeared from the textbooks. However, the earlier need to use every means possible to arrive at a possible crystal structure spawned some pioneering work and led to considerable understanding of the nature of crystals and crystal structures.

The classic work in this area is Kitaigorodskii's *Organic Chemical Crystallography* [6], which contains many still-valuable observations and insights on the nature of intermolecular interactions in molecular crystals. Kitaigorodskii also built a "structure seeker" for the prediction/solution of crystal structures by incorporating the principles of van der Waals radii and closest packing into a mechanical model. In the middle 1960's many of these ideas were incorporated by (the late) Don Williams into his PCK5 program [7], various versions of which he used as an aid for solving the structures of dibenzoylmethane [8], 2,4,6-triphenylverdazyl [9] and others employed in solving the structures of some monosaccharides [10].^{*} A

^{*}A personal note. In 1968 I was a postdoctoral fellow in Ken Trueblood's lab at UCLA. He was working with Don Cram on the structures of paracyclophanes. One of these that came into the lab was the hydrocarbon [2.2]*meta,para* cyclophane, which crystallized in the space group $P2_12_12_1$. In those days (before the birth of MULTAN, SHELX, etc.) such a light atom noncentrosymmetric structure was considered non-routine at best and often not solvable, and it might have been laid to rest. However, the rigid molecule is very similar to [2.2]*para,para* cyclophane which crystallizes in tetragonal space group $P4_2/mmm$, and we had been reinvestigating the structure originally reported by Kathleen Lonsdale [11]. The cell constants of the two structures were also very similar; however, whereas the *para,para* derivative had crystallographic site symmetry *mmm* (a very rare case for molecular crystals [6]), in space group $P2_12_12_1$, $Z'=1$, the *meta,para* derivative was required to have crystallographic site symmetry *1*. The four arrangements possible for the *meta* and *para* rings were generated as input into the Williams PCK5 program at the locations suggested by the tetragonal structure. One of the four produced a significantly lower packing energy than the others as a trial structure, and refined smoothly on data collected manually on a General Electric quarter-circle diffractometer. However, the exocyclic C-C single bonds were approximately 1.62Å, a value much larger than any previously published. Another set of data led to identical results, which we felt could not be published due to this anomalously long bond. Sometime later, Renault *et al.* published another structure of the compound [12, also] in $P2_12_12_1$, but with $Z'=2$, in which 3 out of 4 of the C-C single bonds exceeded 1.54, and subsequently many C-C single bonds exceeding the 'standard' 1.54Å have been reported.

parallel development at the time was Bill Busing's WMIN program [13], which was used by Kvik and Noordik to aid in solving the structure of 2-amino-4-methylpyridine [14].

The field has expanded rapidly over the past thirty years, with an increasing number of practitioners developing a variety of methods. At least three useful summaries with many references to the historically important and current developments have been given by Verwer and Leusen [2], Gavezzotti and Filippini [15] and Price [16]. Price et al. also compiled a useful 'atlas' of organic molecules which have been used in crystal structure prediction studies carried out by lattice energy calculations [17]. Of the ~190 structures reported in this survey 9 are denoted by the authors as having been carried out "in advance of an experimental investigation" – much in the way that traditional predictions would be made.

The title of this paper involves the confluence of two subjects – the calculation of crystal structures on the one hand and polymorphism on the other hand. Some historical developments of the former were noted above. In many ways the beginning of modern systematic study of polymorphism also can be traced to the 1960's with two landmark papers by McCrone [18, 19]. In the first of these McCrone made an oft-quoted provocative statement about the occurrence of polymorphism:

"Every compound has different polymorphic forms, and the number of forms known for a given compound is proportional to the time and energy spent in research on that compound."

In spite of the implied universalism of this statement, there are many very well researched compounds for which polymorphism has not been discovered: naphthalene, sucrose and the pigment perylene red are but a few notable examples. Statistics on the occurrence of polymorphism are hard to come by, because almost any data set is biased, and many instances of polymorphism are not properly reported (this includes false positives as well as false negatives). For instance, a survey of the Cambridge Structural Database on the keywords "polymorph," "form," "phase" and "modification" indicates that about 4% of the included structures are polymorphic. A search on "hydrates and solvates" results in about 23% hits. This survey is biased by the fact that it includes only compounds for which the crystal structures have been reported. Perhaps closer to the other extreme, Steve Byrn recently summarized work from SSCI on over 150 compounds for which >87% had more than one form, 37% were hydrates and 31% were solvates [20]. These statistics may be biased by the fact that some samples at least were known to be polymorphic when received, or special efforts were made to identify multiple crystal forms, even if relatively unstable. Additional statistical surveys have been reported [21].

The general situation is somewhere in between these two sets of statistics: the existence of multiple crystal forms (polymorphs and/or solvates) is not surprising when found, but cannot be foreseen or expected for any compound. Essentially, this has been the state of our knowledge and understanding for many years, and given the increasing interest and importance of multiple crystal forms, it is not surprising that considerable efforts would be made to "predict" the possible existence of polymorphs in particular, and multiple crystal forms in general. One result was the announcement of the "Polymorph Predictor" program in 1994.

4. WHAT WE CAN DO; WHAT WE CAN'T DO

In the introduction to a chapter in a 1997 book entitled *Theoretical Aspects and Computer Modeling of the Molecular Solid State* edited by Angelo Gavezzotti, Robert Gdanitz, one of the authors of the Polymorph Predictor program, quoted three authorities on their view of our ability to predict crystal structures [22]. In a since oft-quoted statement John Maddox, past editor of *Nature*, wrote in 1988 [23]:

"One of the continuing scandals in the physical sciences is that it remains in general impossible to predict the structure of even the simplest crystalline solids."

Similar skepticism was voiced by F. Hawthorne [24]:

"Thus a rigorous general solution to the question of crystal structure prediction may not be forthcoming in the near future."

and Fagan and Ward [25]:

"Unfortunately, the molecular interactions in a lattice become too complicated for any human or computer to handle."

Nevertheless, in the same opening paragraph, Gdanitz [22] concluded that:

"...quite a few articles on predicting molecule crystal structures have been published, showing that *the problem has now become a rather routine task*" (emphasis added).

Perhaps the telltale hint of the author's true estimation of one's predictive ability is the title of the chapter, "*Ab Initio* Prediction of Possible Crystal Structures". The prediction of *possible* crystal structures may be routine, but the prediction of *the* crystal structure clearly is not.

A similarly over optimistic view of the situation may be found in the chapter by Verwer and Leusen [2] in stating that

"...computational methods and algorithms that allow packing simulations with many degrees of freedom,...were not developed until this decade (e.g. the 1990's), making true 'ab initio' crystal structure predictions possible on the basis of molecular information alone."

Possible, perhaps, but the rate of success still leaves a lot to be desired before it can be claimed that the problem has been solved, and crystal structures in general, or polymorphic structures in particular, can be reliably predicted. In reviewing the general situation of the predictability of polymorphs in general, Gavezzotti's emphatic "No" in 1994 and subsequent developments, Dunitz succinctly summarized the situation in 2003. His current answer to the question is [26]:

"...still 'No', although at certain levels of discussion a 'Maybe' or even a conditional 'Yes', may be entertained as possible responses".

Perhaps the greatest service to this effort of developing methods to computationally survey possible crystal structures has been performed by the Cambridge Crystallographic Data Centre, with a series of competition "blind tests." Developers and practitioners of these methods have been invited three times since 1998 to apply their methods to attempting to predict the crystal structures of three compounds on the basis of molecular information alone. The crystal structures of the candidate compounds have been done but not published. The results of two of these blind tests have been published [27, 28]. The rules for the competition say as much about our current ability to computationally predict crystal structures as do the results of the competition. These rules were laid out, "[t]o give a reasonable chance of success within the practical limits of known computer programs...":

- maximum number of atoms including H atoms did not exceed 40;
- space group required to be one of the ten most frequently encountered (i.e. $P2_1/c$, $P-1$, $P2_12_12_1$, $C2/c$, $P2_1$, $Pbca$, $Pna2_1$, Cc , $Pbcn$, $C2$);
- one molecule in the asymmetric unit;"

* The ground rules for the second blind test are quoted here. They differ only slightly from the first.

** Most of the programs used for computing the lattice energy can not take into account the possibility of the molecule lying on a crystallographic special position (Wyckoff position). In a recent survey [29] it was shown that for $Z'=1$ approximately 71% of the molecules lack molecular symmetry, while those that have an inversion center retain that in more than 80% of the cases, those with a rotation axis retain it in approximately 50% of the cases, while those with mirror plane retain that element in about 30% of the cases. Most of these structures with retention of symmetry would not be included in the computed crystal structures since the programs are not capable of handling them. In some algorithms, especially those that compare the calculated powder pattern with an experimental one, the proper spatial arrangement of the molecules might be found, but the space group would not be correctly assigned. Such was the case, for

- no solvent molecules or co-crystals;
- no disorder in the determined structure;
- hydrogen atoms located experimentally in the determined structure.

In addition, the three candidate molecules were categorized by perceived increasing difficulty for prediction:

- rigid molecule with only C, H, N and O atoms, less than 25 atoms;
- rigid molecule with some less common elements (e.g. Br), less than 30 atoms;
- flexible molecule with two degrees of acyclic torsional freedom, less than 40 atoms.

What classes or features of structures are considered too difficult or too challenging for such a competition? "Medium" or "large" molecules (>40 atoms), all of the metal-containing complexes, most of the compounds with less common elements, molecules with more than two conformational degrees of freedom, salts, solvates, and structures with more than one molecule in the asymmetric unit, or molecules lying on special crystallographic positions (inversion center, 2-fold axis, etc.). While the eight allowed space groups comprise approximately 80% of the CSD, by adopting such a restriction one is already reducing one's chance of success by 20%. This is a considerable fraction of the compounds that are of interest to "small molecule" chemists.

What kind of success was achieved for the two blind tests for which results have been published? There were three molecules (but four structures – see below) and 11 participants in the first blind test, for a total of 44 chances of success. Each participant was allowed three choices for each solution, so actually there were 132 possible solutions. There were seven correct predictions, five of them being the first of the three choices. 7/44 is a 16% success rate.

It turned out that the behavior of one of the compounds was particularly relevant to the title of this paper. The compound is a liquid at room temperature, and was crystallized (in orthorhombic *Pbca*) *in situ* at low temperature in a capillary on a diffractometer for the data collection and structure determination, which served as the first structure of reference for the first blind test. A breakdown in the cooling system led to melting, and all subsequent attempts at crystallization yielded a monoclinic *P2₁/c* structure [29,30]. The sequence of events suggests that the monoclinic form is the more stable one, in accord with Ostwald's Rule of Stages, and that the orthorhombic form should be classified as a disappearing polymorph [31]. In terms of the computational prediction of crystal structures, the important point here is that four out of the seven correct responses in the first blind test were for the orthorhombic structure (the less stable form) and if it had not been found initially, the number of correct predictions would be reduced to three (7% success rate). Moreover, if the monoclinic structure had initially crystallized one might never have known about the existence of polymorphs in this system.

Seventeen groups participated in the second blind test, carried out on three structures for a total of 51 possible correct answers. The structures were correctly predicted by two groups for the first molecule, by four groups for the second molecule, with no correct answers for the conformationally flexible third molecule. The success rate is therefore 6/51 or 12%. The participants did conclude that there had been significant progress on the ability to generate crystal structures for rigid molecules, and if the solution were to be found among the top six, rather than the top three, solutions the success rate would rise significantly. The failure of all participants to find a solution for the third molecule in the second round led to additional attempts to prepare a polymorphic structure which might be found among the higher ranked computed ones [32]. The authors determined that the blind test reference structure exhibited an unusual conformation, which made the calculations less likely to succeed. After determining the cell constants of more than 80 crystals they obtained a second polymorph of the material crystallized from nitromethane; however, it crystallizes with two molecules in the asymmetric unit with two different molecular conformations, so that

instance, for the original version of the "Polymorph Predictor", which yielded a solution for benzene in space group *P2₁2₁2₁* (*Z*=4, *Z'*=1, molecules on general positions), when it should have been *Pbca* (*Z*=4, *Z'*=0.5, molecules on inversion centers).

it would not have been included in any of the possible structures for the blind test calculated at the current level of capability.

The example of acetic acid demonstrates one of the major difficulties and challenges still to be overcome. An attempt to computationally predict the crystal structure(s) of acetic acid using both the grid-search method [33] and the Monte Carlo approach [34-36] led to ~100 structures within a narrow energy range of 5 kJ/mol. With various modifications the experimental structure always had a low energy, but it was not possible to distinguish it from the many other solutions. This led the authors to suggest that "other factors need to be considered for genuine structure prediction." In the realm of synthetic organic chemistry, a parallel situation would be to claim that one has carried out the synthesis of the desired product which indeed may be found in the reaction mixture along with ~100 other products, but we don't know how to independently identify or isolate that desired product.

All the methods for calculating lattice energies of possible crystal structures lead to a list that must be ranked. Ideally, the real or possible polymorphic forms appear as the most stable forms in that list. Historically, success has been measured by whether a structure, or polymorphic structures can be properly accounted for, or predicted in that list and subsequently found. I would like to suggest an alternative developmental approach, which I believe has been applied to a much lesser extent, but may be more systematic and fruitful in terms of the long term development of these methods.

The polymorphic systems to be studied should be very well characterized in terms of the relative thermodynamic stability of the various forms, monotropism, enantiotropism, heats of transition, and heats of sublimation, etc., if possible. In the end, the lattice energies that are calculated can and should be benchmarked against those experimental quantities. I do not underestimate the difficulties in obtaining good quality thermodynamic data for all of these quantities, especially sublimation enthalpies, and the paucity of laboratories where such measurements are made. The least demanding benchmark for one of these calculations is that it reproduces the experimentally determined energetic ranking of the various polymorphic forms, without regard to the actual quantities involved. At the next level of stringency, the computed differences in energy between the various polymorphic forms should conform to the experimentally determined differences. As the most demanding test, the computed lattice energies should match the experimentally determined ones. Such a developmental process should result in a force field and search procedure that are at least suitable for a class of functional groups and molecules similar to those of the test molecule or group of molecules. Attempts to develop 'universal' methods or force fields are reminiscent of the search for the Holy Grail.

The computational "proof of concept" experiment would then be to use that force field and those computational methods to generate, *ab initio*, the crystal structures of all of the known polymorphs in the energetic order in which they are known to exist. Suitable (albeit demanding) test systems, might be a compound like ROY, for which seven polymorphs have been identified and characterized [37,38], or *p*'-methylchalcone, for which 13 polymorphic forms have been reported [39] and at least 5 structures have been determined [40].

Table 1
Crystal forms obtained for N,N'-dithiobisphthalimide [from 41]

Form	Solvent for crystallization	Space group	Z'
Solvent-free polymorphs			
(1)	CH ₃ COOC ₂ H ₅	P2 ₁ /c	2
(4)	CH ₃ OH	Pna2 ₁	1
(5)	CH ₃ CN or HCON(CH ₃) ₂	P2 ₁ /c	2
Polymorphs with solvent hydrogen-bonded to (1)			
(6)	CH ₃ NO ₂	P2 ₁ /c	1
Solvent molecules are in isolated cavities			
(7)	C ₆ H ₅ Cl	P $\bar{1}$	1
(8)	C ₆ H ₅ C ₂ H ₅	P2 ₁ /c	2
Solvent molecules are non-localized in channels			
(2)	CH ₂ Cl ₂	C2/c	0.5
(3)	C ₂ H ₅ OH	C2/c	0.5
(9)	C ₆ H ₆ O	C2/c	0.5
(10)	(CH ₃) ₂ COH/H ₂ O	C2/c	0.5
Solvent molecules are localized in channels			
(11)	1,4-(CH ₃) ₂ C ₆ H ₄	P2 ₁ /n	1
(12)	C ₆ H ₅ CH ₃	P2 ₁ /n	1

This process for developing reliable computational methods does not deal with any of the simplifying limiting assumptions made for the blind tests. For crystal structure prediction those will have to be dealt with as well. Some of the challenges are represented by the variety of polymorphs and solvates reported for the relatively simple molecule N,N'-dithiobisphthalimide, summarized in Table 1 [41]. The three solvent-free polymorphs represent a challenge that might be considered possible today, but "predicting" the other four solvated motifs is way beyond our current capabilities.

The description above might leave the impression that computational methods have not progressed sufficiently to provide useful chemical and structural information. As noted at the outset, the contrary is true. In conjunction with a variety of analytical techniques, these methods can provide guidelines and confirming parameters for modeling possible crystal structures. We are still a long way from crystal structure or polymorph *prediction*, but we can learn something about what we might want to look for and obtain structural information when crystal structure analysis is difficult or not possible. The recent case history of acetaminophen illustrates this synergism.

Acetaminophen/paracetamol (e.g. Tylenol®) is the most widely used analgesic and antipyretic which has been known to be dimorphic for many years [42]. The commercially used Form I requires binders for formulation; Form II does not require binders, but transforms to Form I. Considerable effort has gone into improving the solid state properties of acetaminophen, including the search for additional crystal forms that might have improved formulation and stability properties, and these efforts included a computational search for possible new forms. Verwer and Leusen [2] completed one such study with the conclusion that

"Recalculation of the lattice energetic...established that this third structure is actually too unstable to exist."

Chemists often see the prediction of the nonexistence of a substance as a challenge to make it,* and this case was no different. In fact, at the time the statement was made there was already evidence in the

* Philosophers of science have also argued against this approach. For instance, the relevant quotation in a recent book by the father-philosopher/son-scientist, Buddhist monk team: "But not to be able to find something is no proof of its nonexistence." [43].

literature for a third form, detected by microscope hot stage methods [44-46]. Beyer et al. carried out another set of calculations on the possible crystal structure of the compound [47]. Almost simultaneously, a set of experiments based on high throughput crystallizations found evidence for a third form, which was characterized by hot stage microscopy and Raman spectroscopy [48]. The X-ray powder diffraction pattern measured by Peterson et al. [48] also closely matches one of the low energy computed structures of Beyer et al., although the latter had ruled it out as a possible structure on the basis of packing considerations. In any event, the combination of experimental and computational techniques led to significant advances in the characterization and understanding of this system, and it is this synergism, especially between computationally-derived models for crystal structures and powder diffraction [49] which promises to provide the greatest immediate and near term benefit.

5. THE CHALLENGES

What is the current status of our ability to predict crystal structures in general and polymorphs in particular? The situation was recently summarized by S.L. Price, one of the leading practitioners in the field [16]:

“We cannot yet computationally predict the possible polymorphs of any given organic molecule.”

As we have pointed out above, and as Price further notes, the computational studies can provide useful information about polymorphs that we might want to look for. They say nothing about their existence or how they might be prepared. In fact, as Dunitz has noted in the same vein [26]:

“We have a long way to go before we can think of predicting which polymorph will be obtained under any given circumstances.”

From the computational point of view it must be shown that calculated lattice energies can truly reflect or reproduce experimentally determined energetics. This will require much more work on force fields, and the development of computational tools for determining lattice energies. Good thermodynamic data are also required on polymorphic systems to provide the standards against which these calculations can be measured. It is still difficult to distinguish between the global minimum and local minima in computational studies, which means that it is not always possible to computationally determine which polymorphic form is the most stable, and how others may be thermodynamically related to it. Price has also pointed out the necessity for assessing and modeling kinetic factors in the growth of different polymorphs.

In order for the computational techniques to be truly predictive, they must be more comprehensive. Hence, on the more practical level some of the computational challenges that must be met along the way were noted above. (In the following the numbers in parentheses are the current percentages of entries in the CSD). Molecules containing only C,H,N,O comprise about 18.5% of the CSD. Provision must be made for modeling crystals with conformationally flexible molecules, molecules larger than ~40 atoms, crystals of salts, solvates (14.3% of CSD) and hydrates (10.1% of CSD), cocrystals, crystals of organometallic compounds (56%), crystals with more or less than one molecule in the asymmetric unit (35%), crystals in the ‘less common’ space groups (12.7%) – in short the technique must be generalized to cover all the possibilities that are regularly encountered in laboratory situations.

By noting our current limitations Price and Dunitz have touched on some of the exciting challenges facing those studying the chemistry of crystal forms – polymorphs and solvates. I would define them with three rather general questions which we would like to be able to answer for any specific system:

- How many different crystal forms can we expect?
- How do we go about preparing those different crystal forms?
- What will be the properties of those different crystal forms if and when I obtain them?

This contribution made an attempt to assess where we are with regard to the computational tools used to address the first question. There has indeed been significant progress since Kitaigorodskii’s pioneering

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work in the area, and in closing I was reminded of Martin Luther King's inspiring remarks in a memorable speech at the Lincoln Memorial 41 years ago:

"We've come a long way, but we've got a long, long way to go."

6. REFERENCES

- [1] Later referenced [2] as follows: Cerius² User Guide, March 1997, Molecular Simulations, Inc. 9685 Scranton Road, San Diego, CA; <http://www.msi.com/>.
- [2] P. Verwer and F.J.J. Leusen, in *Reviews in Computational Chemistry* 1998, eds. K.P. Lipkowitz and D.B. Boyd (Wiley-VCH, John Wiley and Sons, Inc., New York 1998), p 327.
- [3] A. Gavezzotti, *Acc. Chem. Res.* **27** (1994), 309.
- [4] H. Lipson and W. Cochran, *The Determination of Crystal Structures*, ed. W.L. Bragg, *The Crystalline State*, Vol 3, (Cornell University Press, Ithaca, 1966), Chapter 6.
- [5] C.W. Bunn, *Chemical Crystallography*, (Oxford University Press, Oxford, The Clarendon Press, Oxford, 2nd Edition, 1961), p. 204
- [6] A.I. Kitaigorodskii, *Organic Chemical Crystallography*, (Consultants Bureau, New York, 1961).
- [7] D.E. Williams, *Acta Crystallogr., Sect. A* **25** (1969), 464.
- [8] D.E. Williams, *Acta Crystallogr.*, **21** (1966), 340.
- [9] D.E. Williams, *Acta Crystallogr., Sect. B* **29** (1973), 96.
- [10] P. Zugenmaier and A. Sarko, *Acta Crystallogr., Sect. B* **28** (1972), 3158.
- [11] H. Hope, J. Bernstein and K.N. Trueblood, *Acta Crystallogr. Sect. B* **28** (1972), 1733.
- [12] A. Renault, C. Cohen-Addad, J. Lajzerowicz-Bonnetau, J.P. Dutasta and M.J. Crisp, *Acta Crystallogr., Sect. B* **38** (1987), 480.
- [13] W.R. Busing, *Acta Crystallogr., Sect. A* **28** (1972), S252.
- [14] Å. Kvik and J.H. Noordik, *Acta Crystallogr., Sect. B* **38** (1977), 2862.
- [15] A. Gavezzotti and G. Filippini, in *Theoretical Aspects and Computer Modeling of the Molecular Solid State* 1997, ed. A. Gavezzotti (John Wiley and Sons, Inc., New York 1997), p 61.
- [16] S.L. Price, *Adv. Drug Delivery Revs.*, **56** (2004), 301.
- [17] T. Beyer, T. Lewis and S. Price, *Cryst. Eng. Comm.* **44** (2001), 1.
- [18] W.C. McCrone, in *Physics and Chemistry of the Organic Solid State*, 1965 eds. D. Fox, M.M. Labes and A. Weisberger (Wiley Interscience, New York 1965), p 725.
- [19] J. Halebian and W.C. McCrone, *J. Pharm. Sci.* **58** (1969), 911.
- [20] S.R. Byrn, presented at the 35th Erice School on Crystallography, "Diversity Amidst Similarity: A Unified Approach to Polymorphs, Solvates and Phase Changes", Erice, Sicily, June, 2004.
- [21] J.A.R.P. Sarma and G.R. Desiraju, in *Crystal Engineering. The Design and Applications of Functional Solids*, eds. K.R. Seddon and M. Zaworotko (Klewer, Dordrecht 1999), pp. 325-356; U.J. Griesser and A. Burger, "Statistical Aspects of the Occurrence of Crystal Forms Among Organic Drug Substances" Poster P09.013 at the International Union of Crystallography Congress and General Assembly, Glasgow, Scotland, Aug. 4-13, 1999. Book of Abstracts, p. 533; U.J. Griesser, *Acta Crystallogr., Sect. A* **58** Supplement (2002), C241.
- [22] R.J. Gdanitz, in *Theoretical Aspects and Computer Modeling of the Molecular Solid State* 1997, ed. A. Gavezzotti (John Wiley and Sons, Inc., New York 1997), p 18.
- [23] J. Maddox, *Nature*, **335** (1988), 201.
- [24] F.C. Hawthorne, *Nature*, **345** (1990), 297.
- [25] P.J. Fagan and M.D. Ward, *Scientific American*, **267**(1) (1992), 28.
- [26] J.D. Dunitz, *Chem. Comm.*, (2003), 545.

- [27] J.P.M. Lommerse, W.D.S. Motherwell, H.L. Ammon, J.D. Dunitz, A. Gavezzotti, D.W.M. Hofmann, F.J.J. Leusen, W.T.M. Mooij, S.L. Price, B. Schweizer, M.U. Schmidt, B.P. van Eijck, P. Verwer and D.E. Williams, *Acta Crystallogr., Sect. B* **56** (2000), 697.
- [28] W.D.S. Motherwell, J.P.M. Lommerse, H.L. Ammon, J.D. Dunitz, A. Dzyabchenko, P. Erk, A. Gavezzotti, D.W.M. Hofmann, F.J.J. Leusen, J.P.M. Lommerse, W.T.M. Mooij, S.L. Price, H. Scheraga, B. Schweizer, M.U. Schmidt, B.P. van Eijck, P. Verwer and D.E. Williams, *Acta Crystallogr., Sect. B* **58** (2002), 647.
- [29] E. Pidcock, W.D.S. Motherwell and J.C. Cole, *Acta Crystallogr., Sect. B* **59** (2003), 634.
- [30] R. Boese, private communication.
- [31] J.D. Dunitz and J. Bernstein, *Acc. Chem. Res.* **28** (1995), 193.
- [32] R.K.R. Jetti, R. Boese, J.A.R.P. Sarma, L.S. Reddy, P. Vishweshwar and G.R. Desiraju, *Angew. Chem. Int. Ed.* **42** (2003), 1963.
- [33] W.T.M. Mooij, B.P. van Eijck, S.L. Price, P. Verwer and J. Kroon, *J. Comput. Chem.* **19** (1998), 459.
- [34] H.R. Karfunkel and F.J.J. Leusen, *Speedup* **6** (1992), 43.
- [35] H.R. Karfunkel, F.J.J. Leusen and R.J. Gdanitz, *J. Comput.-Aid Mat. Des.*, **1** (1993), 177.
- [36] H.R. Karfunkel, and R.J. Gdanitz, *J. Comput. Chem.*, **13** (1992), 1171.
- [37] L. Yu, G.A. Stephenson, C.A. Mitchell, C.A. Bunnell, S.V. Snorek, J.J. Bowyer, T.B. Borchardt, J.G. Stowell and S.R. Byrn, *J. Am. Chem. Soc.* **122** (2000), 585.
- [38] L. Yu, *J. Phys. Chem. A* **106** (2002), 544.
- [39] C. Weygand and H. Baumgärtel, *Lieb. Ann.* **469**, (1929) 251.
- [40] J. Bernstein and J.-O. Henck, *Cryst. Eng.* **1** (1998), 119.
- [41] D.M.M. Farrell, C. Glidewell, J.N. Low, J.M.S. Skakle and C.M. Zakaria, *Acta Crystallogr., Sect. B* **58** (2002), 289.
- [42] G. Nichols and C.S. Frampton, *J. Pharm. Sci.* **87** (1998), 684.
- [43] J.-F. Revel and M. Ricard, *The Monk and the Philosopher*, (Thorsons, London, 1998), p. 57.
- [44] A. Burger, *Acta Pharm. Technol.* **28** (1982), 1.
- [45] P. Di Martino, P. Conflant, M. Drache, J.-P. Huvenne and A.-M. Guyot-Hermann, *J. Therm. Anal.* **48** (1997), 447.
- [46] M. Szelagiewicz, *Therm. Anal.* **57** (1999), 23.
- [47] T. Beyer, G.M. Day and S.L. Price *J. Am. Chem. Soc.* **123** (2001), 5086.
- [48] M.L. Peterson, S.L. Morissette, C. McNulty, A. Goldsweig, P. Shaw, M. LeQuesne, J. Monagle, N. Encina, J. Marchionna, A. Johnson, J. Gonzalez-Zugasti, A.V. Lemmo, S.J. Ellis, M.J. Cima and Ö. Almarsson, *J. Am. Chem. Soc.* **124** (2002), 10958.
- [49] W.I.F. David, K. Shankland, L. McCusker and Ch. Baerlocher, (eds.) *Structure Determination from Powder Diffraction Data* (Oxford University Press, Oxford, 2000).

Expanding the Scope of Crystal Form Evaluation in Pharmaceutical Science

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ABSTRACT - The commentary seeks to provide a brief history and perspective on the importance of crystal forms of pharmaceuticals as a means of achieving performance criteria. The expanding scope of crystal form selection, emergence of crystal engineering in pharmaceutical science and pharmaceutical co-crystals are topics of this brief review.

THE ROLE OF MOLECULAR CRYSTALS IN PHARMACEUTICAL SCIENCE

Chemists and engineers in the pharmaceutical industry generally seek to deliver crystalline forms of their active compounds, mainly due to the inherent stability of crystalline materials and the well-established impact of crystallization processes on purification and isolation of chemical substances (1). Increasing attention is now being paid to the impact of materials properties in drug discovery and early development (2) as the drug compounds tend to be very valuable materials. The pharmaceutical industry's mission for the material is to rapidly advance development programs with good confidence that form and formulation problems are unlikely to arise and to maximize a compounds' potential as a therapeutic. This commentary seeks to raise the profile of crystal form studies and the emerging topic of crystal engineering in pharmaceutical science by discussing the "state-of-the-art" relating to pharmaceutical crystals and by expanding on possibilities that exist for future developments. In keeping with the common goal of making better products, faster and cheaper, we propose a paradigm of pro-active material design in pharmaceutical research. Rather than settling for the

physical forms that the pure compounds intrinsically display, we should be aiming to identify the physical / material properties required of the target drug and make crystalline forms to meet those needs. We are not yet actively engaging in crystal engineering in the industry, and therefore we pose the question: *How can we proceed toward making the crystal form that we want to use?*

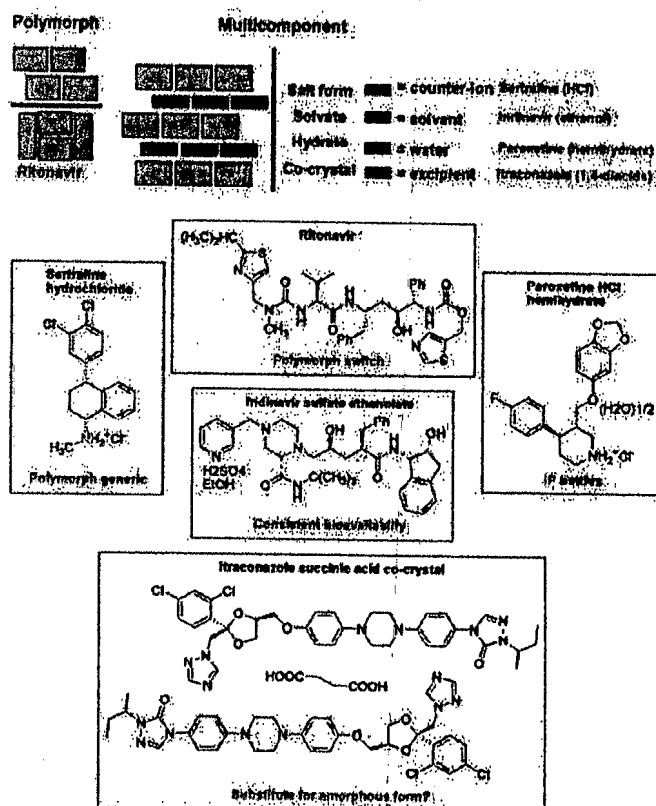


Figure 1. Examples of the types of crystal forms of pharmaceutical compounds used in products along with some issues and opportunities associated with each example.

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Polymorphism and pharmaceuticals

The phenomenon of crystal polymorphism, where the same chemical compound exists in more than one unique crystalline form, has been appreciated for over a century (3,4). Polymorphs of pharmaceuticals and drug candidates can occur in all types of phases (pure compounds, salts, solvates; for examples see Figure 1), though there is no way to predict the practical extent of polymorphism of any given compound (5). Based on recent reviews and commentaries, many strides have been taken towards better understanding of crystal polymorphism of pharmaceuticals (6,7,8). According to lore and some published examples, the puzzling and unpredictable phenomenon of crystal polymorphism has affected many projects in pharmaceutical research and development over the last few decades. Although most accounts of polymorphic transformation during product development remain anecdotal, there exist published examples of the problem from both early stage compounds (9) and marketed drugs (10).

The impact of crystal polymorphism and solvation state on pharmaceutical product value has been illustrated by costly product failures and protracted patent litigation examples. The former was most notably exemplified by the Norvir® capsule product failure in 1998, which was recounted and rationalized by solving the crystal structures of the Ritonavir polymorphs (10). In theory, all pharmaceuticals are vulnerable to such an unexpected and unlucky event which in the case of Ritonavir resulted in enormous cost and inconvenience for the innovator and impacted the patients' use of this important HIV drug for roughly one year when no capsule formulation was available. Recently a new, meta-stable form of 5-fluorouracil was reported highlighting the potential for even very old drugs to unexpectedly reveal new polymorphs (11). On the litigation side, early entry of generic versions of some notable drugs has been enabled based on the use of a patently distinct but pharmaceutically equivalent polymorphs (or hydrates) (12). Ranitidine HCl (Zantac®) (5) and Paroxetine HCl hemihydrate (Paxil®) (13) are but two prominent examples where patent strategy and gaming created significant uncertainty about the value of a drug franchise. In response to these challenges, the pharmaceutical industry has developed processes and techniques for the

identification and characterization of polymorphs and solvates of compounds of interest (6,14). Academics are contributing with both experimental and theoretical insights into crystal form study, prediction and engineering (7). Regulatory agencies also joined the effort in the 1990's, as exemplified by the decision-tree approach to evaluating the impact of polymorphism on product performance (15,16). It seems fair to conclude that developments in the late 1980's and many more during the 1990's have brought the industry's level of awareness, detection capability and decision making regarding polymorphs and solvates to new heights.

Beyond crystals and polymorphs

Another phase type of interest in pharmaceutical systems is the amorphous state of a compound. Amorphous drug preparations of small molecule drugs exist in the marketplace (17), and they usually represent deliberate efforts to avoid crystalline forms in order to meet a delivery objective. This is done when a crystalline form is unsuitable for oral absorption due to insufficient aqueous solubility and inadequate dissolution rate, particularly when particle size reduction does not ameliorate the impact of the latter to a satisfactory degree. Infrequently, however, no suitable crystalline forms of a compound can be found making the amorphous material the only viable option. Examples include itraconazole, quinapril HCl, zafirlukast, cilastatin and nelfinavir mesylate. In such cases, it would seem important to gain insights into the structural factors that lead to the outcome, in an effort to assess risk of eventual crystallization events. The characterization and application of amorphous pharmaceutical compounds and dispersions have been topics of review elsewhere, and will not be further discussed, given that the focus of the present contribution is on crystalline compounds (18-20).

Particle size reduction of poorly absorbed compounds is a proven approach to enhance biopharmaceutical performance. Milling to progressively smaller sizes can be considered a progressive approach towards amorphous, high-energy states. A special case of such crystal size modification is the advent of the nano-dispersion approach in pharmaceutical products. A key technology development in this regard is

NanoCrystals® (Elan corp.), which has been successfully applied in three oral products to date: Rapamune®, Tricor® and Emend®. The latter two compounds are excellent examples of the impact of media milling and physically stabilizing a crystalline dispersion of nanometer dimensions (100's of nm) particles with the ultimate goal of significant bioavailability improvement. The physical challenge is one of re-growth of particles (Ostwald ripening), which is addressed by the use of specific surface stabilizers in formulation development. The active ingredient in Tricor, fenofibrate, is a low-melting, oil soluble (but water-insoluble) substance. The bioavailability of this compound is increased by decreasing particle size, with the nano-dispersion being close to optimal. In addition, the NanoCrystal formulation of Tricor shows a decreased dependence of oral bioavailability on food compared to the previous formulation of Tricor (21). In the case of Emend, the solubility of the drug substance, aprepitant, is on the order of 0.3 µg/mL and the doses are in 100's of mg/day. Oral bioavailability of this drug is a strong function of the particle size, with the smallest achievable crystals being optimal (22). The example illustrates that size modification is a viable strategy to be considered alongside form modification.

Salt selection – A powerful strategy for crystal form optimization

Pharmaceutical developers have focused efforts on finding and formulating a thermodynamically stable crystalline form with acceptable physical properties for a given compound. This is reasonable, given the need to avoid cascading from a meta-stable form to a more stable one in unpredictable fashion. Occasionally certain physical properties, such as low aqueous solubility, are limiting to performance of the compound, leading to poor oral bioavailability or insufficient solubility for an injection formulation. One of the main strategies used to affect physical performance of a compound and one that is often employed by pharmaceutical scientists is the practice of salt selection (23). At least half of compounds in marketed products are in the form of a salt for one reason or another. This fact alone speaks to the versatility of the salt selection approach. Salt forms of a pharmaceutical can have many benefits, such as improved stability

characteristics, optimal bioavailability and aqueous solubility for an injectable formulation. Salts, like all other crystalline forms, are subject to polymorphism and solvate formation, thus requiring the same form identification studies as are needed for a neutral compound. A remarkable example of co-optimization of properties is indinavir (HIV protease inhibitor), which is marketed as the sulfate salt ethanol solvate (24,25). The crystalline free base has variable oral bioavailability in dogs (26,27) and humans (28). While acidic solutions of the base compound showed good oral pharmacokinetics, the stability of the drug in acidic solution is not consistent with a product (26). Therefore, the discovery of the salt form ensured both shelf stability and robust bioavailability performance.

The salt selection strategy is limited in two ways. First, salt formation relies on the presence of one or more ionizable functional groups in the molecule; many drugs and development compounds lack this feature. Second, our ability to predict *a priori* whether a given compound will form a *crystalline* salt (or salts) is non-existent. The ability to actively identify crystalline salt forms has been confined to manual empirical evaluation using multiple salt formers for a given acid or base. Recently advances have been made in the area of high-throughput salt selection and crystal engineering strategies associated with salt formation (14,29-32). In one case, we have advocated the simultaneous assessment of polymorphism as a way to help rank the developability of different crystalline salts (14). While salt forms will continue to have a prominent place in pharmaceutical science, the need for enhanced productivity dictates that every advantage must be sought to aid the design of an appropriate crystalline form of an active molecule. Specifically, the ability to design scaffolds into crystalline forms will enhance our capacity to convert interesting molecules into effective drugs. Crystal engineering offers some additional tools in this regard.

Crystal engineering and co-crystals

Crystal engineering is generally considered to be the design and growth of crystalline molecular solids with the aim of impacting material properties. A principal tool is the hydrogen bond, which is responsible for the majority of directed

intermolecular interactions in molecular solids. Co-crystals are multi-component crystals based on hydrogen bonding interactions without the transfer of hydrogen ions to form salts – this is an important feature, since Brønsted acid-base chemistry is not a requirement for the formation of a co-crystal. Co-crystallization is a manifestation of directed self-assembly of different components. Co-crystals have been described of various organic substances over the years (33,34) and given various names, such as addition compounds (35,36) molecular complexes (37,38) and heteromolecular co-crystals (39). Regardless of naming convention, the essential meaning is that of a multi-component crystal where no covalent chemical modification of the constituents occurs as a result of the crystal formation. Pharmaceuticals co-crystals have only recently been discussed as useful materials for drug products.

Pharmaceutical co-crystals

Pharmaceutical co-crystals can be defined as crystalline materials comprised of an active pharmaceutical ingredient (API) and one or more unique co-crystal formers, which are solids at room temperature. Co-crystals can be constructed through several types of interaction, including hydrogen bonding, π -stacking, and van der Waals forces. Solvates and hydrates of the API are not considered to be co-crystals by this definition. However, co-crystals may include one or more solvent/water molecules in the crystal lattice.

An example of putative design, a construction and preparation process is shown in Figure 2 for the 5-fluorouracil:urea 1:1 co-crystal(40). This real example neatly illustrates the opportunity and challenge that exists currently with designing pharmaceutical co-crystals. Firstly, the 'design' is challenging because we have no ability to predict the exact crystal structure that may result from a crystallization attempt. By analogy to the challenge of deriving protein structure from first principles, the primary sequence (chemical structure in our case) is known and elements of secondary structure (the 2-D tape construction in Figure 2) are somewhat discernible from primary information. Prediction of the actual 3-D folded conformation (tertiary structure or obtained by self-assembly) is not possible. In other words, while we currently

have the ability to project which things associate in what approximate manner on the secondary level, crystal structure prediction is essentially an intractable proposition. By extension, and just as the exact function of a protein and quantitative parameters of activity are not predictable from primary and secondary structure, the prediction of crystal properties is not possible in the absence of structural information and measurements.

There is early evidence that practitioners were aware that apparent co-crystallization of drugs could lead to useful preparations (41). In fact, a 'chemical compound' composed of sulfathiazole and proflavin dubbed flavazole was used to treat bacterial infection during the Second World War (42). The case of flavazole reveals insight into how two different molecules might interact in a putative co-crystal: "... *flavazole is definitely a chemical compound containing equimolar proportions of sulphathiazole and proflavin base. It is believed that combination occurs through the acidic sulphonamide group (SO_2NH) of the sulphathiazole and the basic centres of the proflavin. Perhaps the most realistic expression of the formula would be to place proflavin and sulphathiazole side by side with a comma between them.*" (42) In the second half of the 20th century, interest in co-crystals evolved into the directed study of intermolecular interactions in crystalline solids (43-45). The technical development of routine single-crystal structure determination led to a watershed of data, now largely accessible through the Cambridge Structural Database (CSD) (46,47). The structural data have become useful for understanding the intermolecular interactions in co-crystals in atomic level detail (48). Using insight gained from analysis of the CSD and directed experimentation, scientists attempt design of co-crystals with specific properties, such as color or non-linear optical response, by selecting starting components with appropriate molecular properties likely to exhibit specific intermolecular interactions in a crystal (49-52). However, even when chemically compatible functional groups are present it is not possible to accurately predict if a co-crystal, a eutectic mixture or simply a physical mixture will result from any given experiment. As a result of these complexities, attention has been directed at the identification and characterization of intermolecular packing motifs with the goal of developing principles for co-crystal materials (53).

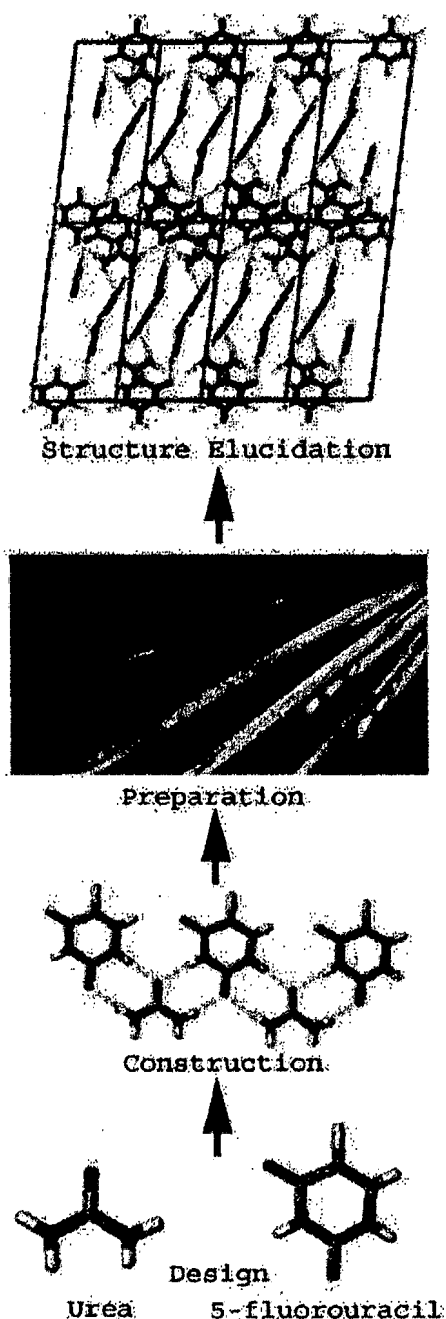


Figure 2. Steps involved in crystal engineering of a pharmaceutical phase, exemplified by the real example of co-crystallization of 5-fluorouracil and urea.

PROSPECTS FOR CRYSTAL ENGINEERING AND PHARMACEUTICAL CO-CRYSTALS

At the beginning of the 21st century, the field of crystal engineering has experienced significant development. Importantly, crystal engineering principles are now being actively considered for application to pharmaceuticals to modulate the properties of these valuable materials (54). Because the physical properties that influence the performance of pharmaceutical solids are reasonably well appreciated, there is a unique opportunity to apply crystal engineering techniques and the appropriate follow-up studies to solve real world problems, such as poor physical and chemical stability or inadequate dissolution for appropriate biopharmaceutical performance of an oral drug. As structures and series of pharmaceutical co-crystals have begun to appear, we again find that properties cannot be predicted from the structures. Nevertheless, occasional trends have been suggested. For example, insoluble drug compounds co-crystallized with highly water soluble complements tend to achieve kinetic solubilities in aqueous media several times greater than the pure form (55,56). There are also more possible phases for each given active compound to consider, thus there will arguably be a greater opportunities for property enhancement. In terms of stability enhancement and solubilization, the example of the series of itraconazole co-crystals with pharmaceutically acceptable 1,4-diacids (55) suggests a strategy alternative to amorphous drug formulation. The co-crystal options presented retain the stability inherent in a crystalline state, while allowing for solubilization that significantly exceeds that of crystalline itraconazole base and rivals the performance of the engineered amorphous bead formulation (Sporanox®).

Where are we now? From recent literature it appears that knowledge gained over the past century and increasingly sophisticated screening techniques developed within the last decade are paving the way towards design of co-crystals with potentially improved pharmaceutical properties (55-58). In terms of the application to pharmaceutical systems, the field of crystal engineering is developing the retro-synthetic understanding of crystal structure using reasoning that is analogous to that applied by organic chemists. For example, the retro-synthetic approach in covalent synthesis

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operates on the level of a single molecule, while the analogous effort in crystal engineering focuses on the "supermolecule": The assemblies that define the crystalline arrangement of the molecules as they self-organize into the solid-state. The parallels between the development of crystal engineering and synthetic organic chemistry run still deeper. Methodologies for carrying out these crystallizations are being developed alongside the development of new robust motifs (6,53,55,57,60). The importance of the solubility and dissolution relationships of the components of a putative co-crystal is becoming a matter of significant investigation (56,60). The same can be said for the roles of additives in templating novel forms. Mechanical milling of materials has also been documented as a means to make co-crystals, and a recent example of polymorphic forms of caffeine:glutaric acid illustrates the opportunities of this type of processing to influence crystal form (61). With an increase in the understanding of the modes of self-assembly, one can start to address the design aspect towards making pharmaceutical co-crystals.

There remain several limitations to the application of what is currently known to the design of useful materials. As mentioned earlier, it remains intractable to reliably predict crystal structure. Multi-component crystals are well out of reach for prediction due in part to complex energetic landscapes, lack of appropriate charge density models and a large number of degrees of freedom, making computation unfeasible. Moreover, there is only a qualitative understanding of the interplay between intermolecular interactions and materials performance, especially for properties relevant to pharmaceuticals such as solubility, dissolution profile, hygroscopicity and melting point. But the saving grace of the co-crystal approach comes in two guises: Complementarity and diversity. On the topic of complementarity, it is possible, by way of CSD database mining for instance, to identify trends of hetero-synthron occurrence in model systems. As for the diversity aspect, the space of possible co-crystal formers is large, limited only by pharmaceutical acceptability. Coupled with parameters such as stoichiometry variation and increase in the number of components (binary systems can be expanded into ternary ones, etc.), the opportunities appear vast.

THE FUTURE OF CRYSTAL ENGINEERING IN PHARMACEUTICAL SCIENCE

Where are we going? At this point, we have only just scratched the surface of materials science-driven pharmaceutical product design. In the 21st century, practitioners of pharmaceutical chemistry need to enumerate and exploit the opportunities of crystal form design that nature affords us, and thus gain increasing ability to design the materials we need from the molecules that we seek to convert into pharmaceuticals. Learning will be facilitated by advances in crystallization automation (6,62), microscopy-spectroscopy techniques (Raman and IR microscopy) and new techniques such as terahertz spectroscopy and AFM, along with increasingly sophisticated X-ray diffraction lab instrumentation. In addition, further enhancements in the data mining tools associated with the CSD operating on an ever increasing number of high-quality crystal structures will undoubtedly lead to new knowledge and principles of interaction.

The challenge placed before pharmaceutical scientists, now and in the future, is the following: (i) to understand the requirement of a particular compound in terms of materials structure and properties, and (ii) to creatively integrate crystal engineering within the limits of pharmaceutical acceptability of components to obtain new forms of active ingredients with desirable properties for formulation and delivery. It should become the collective mantra of medicinal chemists, process engineers and pharmaceutical scientists to "design and make the material we need." This mantra can form the common aspiration for an industry that is in significant need of innovation and productivity enhancement.

CONCLUSIONS

The rapidly evolving field of pharmaceutical crystal engineering is primed for a prominent position in the development and improvement of pharmaceutical products. Scientists in the pharmaceutical industry need to be attuned to the possibilities that the approach can offer. In the next few decades, this awareness needs to become general across the industry and among the disciplines that contribute to drug development. The

expanding role of crystal engineering in pharmaceutical science is waiting to be filled with insights and experience from past work in other areas, stimulated by present challenges in the interest of developing the drugs of the future.

REFERENCES

- [1] E. L Paul, H.-H. Tung, and M. Midler. Organic crystallization processes. *Powder Technology*, 150:133-143, 2005.
- [2] C. R. Gardner, C. T. Walsh and Ö. Almarsson. Drugs as materials: Valuing physical form in drug discovery, *Nature Reviews Drug Disc*, 926-934, 2004.
- [3] W. Ostwald. Studien über die Bildung und Umwandlung fester Körper. *Z Phys Chem*, 22:289, 1897.
- [4] L. Kofler and A. Kofler. Thermo-mikro-methoden zur Kennzeichnung organischer Stoffe und stoffgemische. Innsbruck, Wagner, 1954.
- [5] Bernstein J., *Polymorphism in Molecular Crystals*. Oxford University Press: New York, New York, 2002.
- [6] S. L. Morissette, Ö. Almarsson, M. L. Peterson, J. Remenar, M. Read, A. Lemmo, S. Ellis, M. J. Cima and C. R. Gardner. High-throughput Crystallization: Polymorphs, Salts, Co-crystals and Solvates of Pharmaceutical Solids. *Adv Drug Deliv Rev B*, 275-300, 2004.
- [7] D. Sharmistha and D. J. Grant. Crystal Structures of Drugs: Advances in Determination, Prediction and Engineering. *Nature Reviews Drug Discovery*, 3: 42-57, 2004.
- [8] D. Singhal and W. Curatolo. Drug Polymorphism and dosage form design: a practical perspective. *Adv Drug Deliv Rev*, 56: 335-347, 2004.
- [9] N. Lewis. Shedding Some Light on Crystallization Issues: Lecture Transcript for the First international Symposium on Aspects of Polymorphism and Crystallization – Chemical Development Issues. *Org Proc Res Dev*, 4: 407-412, 2000.
- [10] J. Bauer, S. Spanton, R. Henry, J. Quick, W. Dziki, W. Porter and J. Morris. Ritonavir: An Extraordinary Example of Conformational Polymorphism. *Pharm Res*, 18: 859-866, 2001.
- [11] A. T. Hulme, S. L. Price and D. A. Tocher. A New Polymorph of 5-Fluorouracil Found Following Computational Crystal Structure Predictions. *J Am Chem Soc*, 127:1116-1117, 2005.
- [12] J. Lucas and P. Burgess. When Form Equals Substance: The Value of Form Screening in Product Life-Cycle Management. *Pharma Voice*, 2004.
- [13] J. Lucas and P. Burgess. The Paxil Patent: Four Simple Words, One Complex Claim Construction Case. *Pharmaceutical Law & Industry*, 2:2004.
- [14] J. F. Remenar, J. M. MacPhee, B. K. Larson, V. A. Tyagi, J. Ho, D. A. McIlroy, M. B. Hickey, P. B. Shaw and Ö. Almarsson. Salt selection and simultaneous polymorphism assessment *via* high-throughput crystallization: the case of sertraline, *Org Proc Res Dev*, 7:990-996, 2003.
- [15] S. R. Byrn, R. R. Pfeiffer, M. Ganey, C. Hoiberg, and G. Poochikian. Pharmaceutical Solids: A strategic approach to regulatory considerations. *Pharmaceutical Research*, 12:945, 1995.
- [16] ICH Steering Committee. 11-10-2000. Good Manufacturing Practice Guide For Active Pharmaceutical Ingredients Q7a, ICH Harmonized Tripartite Guideline.
- [17] Ö. Almarsson and C. R. Gardner. Novel approaches the issues of developability. *Current Drug Discovery*, January:21-26, 2003.
- [18] B. C. Hancock and G. Zografi. Characteristics and significance of the amorphous state in pharmaceutical systems. *J Pharm Sci*, 86:1-12, 1997.
- [19] A. R. M. Serajuddin. Solid Dispersion of Poorly Water-Soluble Drugs: Early Promises, Subsequent Problems, and Recent Breakthroughs. *J Pharm Sci*, 88:1058- 1066, 1999.

- [20] L.Yu. Amorphous pharmaceutical solids: preparation, characterization and stabilization. *Adv Drug Deliv Rev*, 48:27-42, 2001.
- [21] See www.tricor.com
- [22] Y. Wu, A. Loper, E. Landis, I. Hettrick, L. Novak, K. Lynn, C. Chen, K. Thompson, R. Higgins, S. Shelukar, G. Kwei and D. Storey. The role of biopharmaceutics in the development of a clinical nanoparticle formulation of MK-0869: a Beagle dog model predicts improved bioavailability and diminished food effect on absorption in human. *Int J Pharm*, 285:135-46, 2004.
- [23] P. H. Stahl and M. Nakano Pharmaceutical Aspects of the Drug Salt Form. *Handbook of Pharmaceutical Salts: Properties, Selection, and Use*. New York: Wiley-VCH/VCHA, 2002.
- [24] J. H. Lin, D. Ostovic and J. P. Vacca, The Integration of Medicinal Chemistry, Drug Metabolism, and Pharmaceutical Research and Development in Drug Discovery and Development, chapter 11: The Story of Crixivan®, and HIV Protease Inhibitor, in *Integration of Pharmaceutical Discovery and Development* (ed. Borchardt et al.), Plenum Press, New York 1998.
- [25] B. D. Johnson, A. Howard, R. Varsolona, J. McCauley and D. K. Ellison. Indinavir Sulfate, in Harry G. Brittain (ed), *Analytical Profiles of Drug Substances and Excipients*, Academic Press: San Diego, 1999; V26, pp. 319-357.
- [26] G.Y. Kwei, L. B. Novak, L. A. Hettrick, E. R. Reiss, D. Ostovic, A. E. Loper, C. Y. Lui, R. J. Higgins, I. W. Chen, J. H. and Lin. , Rediospecific Intestinal Absorption of HIV protease inhibitor L-735,524 in beagle dogs. *Pharm Res*, 12:884, 1995.
- [27] J. H. Lin, I.-W. Chen, K. J. Vastag, and D. Ostovic. pH-dependent oral absorption of L-735,524, a potent HIV protease inhibitor, in rats and dogs. *Drug Metab Disp* 23:730-735, 1995.
- [28] K. C. Yeh, P. J. Deutsch, H. Haddix, M. Hesney, V. Hoagland, W. D. Ju, S. J. Justice, B. Osborne, A. T. Sterrett, J. A. Stone, E. Woolf and S. Waldman. Single-Dose Pharmacokinetics of Indinavir and the Effect of Food. *Antimicrob. Agents Chemother*, 42:332, 1998.
- [29] P. J. Desrosiers. The potential of preform. *Modern Drug Discovery*, 7:40-43, 2004.
- [30] E. A. Collier. A crystallization / crystal engineering approach to aid salt selection – anions. UMIST – Institute of Science and Technology, Dept. of Chem. Eng. 2004.
- [31] O. Félix, M. W. Hosseini, A. De Cian and J. Fischer. Crystal engineering of 2-D hydrogen bonded molecular networks based on the self-assembly of anionic and cationic modules. *Chem Commun*, 281-282, 2000.
- [32] C. B. Aakeroy and M. Niewenhuzen. Hydrogen-bonded layers of hydrogen malate anions – a framework for crystal engineering. *J Am Chem Soc*, 116:10983-10991, 1994.
- [33] For example, d-glucose:sodium chloride monohydrate is described in F. v. Kobell and J. F. *Prakt Chemie*, 28:489, 1843.
- [34] Quinhydrone was described in F. Wöhler. *Untersuchungen über das Chinon. Annalen*, 51:153, 1844.
- [35] For example, A. Buguet. Cryoscopy of Organic Mixtures and Addition Compounds. *Compt Rend*, 149:857-8, 1910.
- [36] H. Grossmann. Thiourea. *Chemiker-Zeitung*, 31:1195-6, 1908.
- [37] For example see, A. Damiani, P. De Santis, E. Giglio, A. M. Liquori, R. Puliti and A. Ripamonti. The crystal structure of the 1:1 molecular complex between 1,3,7,9-tetramethyluric acid and pyrene. *Acta Crystallogr*, 19:340-8, 1965.
- [38] J. N. Van Niekerk and D. H. Saunder. The crystal structure of the molecular complex of 4,4'-dinitrobiphenyl with biphenyl. *Acta Crystallogr*, 1:44, 1948.
- [39] S. Pekker, E. Kovats, G. Oszlanyi, G. Benyei, G. Klupp, G. Bortel, I. Jalsovszky, E. Jakab, F. Borondics, K. Kamaras, M. Bokor, G. Kriza, K. Tompa and G. Faigel. Rotor-stator molecular crystals of fullerenes with cubane. *Nature Materials*, 4:764-767, 2005.
- [40] 5-Fluorouracil:urea. $C_4H_3N_2O_2F.CH_4N_2O$; C2/c; $a = 9.461(3) \text{ \AA}$, $b = 10.487(3) \text{ \AA}$, $c = 15.808(4)$

- λ , β = 99.89(7)°; Z = 4; T = 100(2) K; GOF = 1.023, R^2 = 0.0663; wR^2 = 0.1753.
- [41] C. G. Santesson. Addition Combinations. *Archiv fuer Experimentelle Pathologie und Pharmacologie*, 118:313-24, 1926.
- [42] J. McIntosh, R. H. M. Robinson, F. R. Selbie, J. P. Reidy, H. Elliot Blake and L. Guttman. *Lancet*, 249:97-99, 1945.
- [43] K. Hoogsteen. The crystal and molecular structure of a hydrogen-bonded complex between 1-methylthymine and 9-methyladenine. *Acta Crystallogr*, 16:907-16, 1963.
- [44] F. S. Mathews, and A. Rich. The molecular structure of a hydrogen-bonded complex of N-ethyladenine and N-methyluracil. *J Mol Bio*, 8(1):89-95, 1964.
- [45] H. M. Sobell, K. Tomita and A. Rich. The crystal structure of an intermolecular complex containing a guanine and a cytosine derivative. *Proc Natl Acad Sci USA*, 49:885-92, 1963.
- [46] F. H. Allen. The Cambridge Structural Database: A quarter of a million crystal structures and rising. *Acta Crystallogr*, B58:380-388, 2002.
- [47] I. J. Bruno, J. C. Cole, P. R. Edgington, M. Kessler, C. F. Macrae, P. McCabe, J. Pearson and R. Taylor. New software for searching the Cambridge Structural Database and visualizing crystal structures. *Acta Crystallogr*, B58:389-397, 2002.
- [48] F. H. Allen, O. Kennard and R. Taylor. Systematic analysis of structural data as a research technique in organic chemistry *Acc Chem Res*, 16:146-153, 1983.
- [49] M. C. Etter. A new role for hydrogen-bond acceptors in influencing packing patterns of carboxylic acids and amides. *J Am Chem Soc*, 104:1095-6, 1982.
- [50] M. C. Etter and G. M. Frankenbach. Hydrogen-bond directed cocrystallization as a tool for designing acentric organic solids. *Chem Mater*, 1:10-12, 1989.
- [51] Desiraju, G. R., *Crystal Engineering. The Design of Organic Solids*, Materials Science Monographs 54, Elsevier, Amsterdam, 1989.
- [52] J. A. R. P. Sarma and G. R. Desiraju. Crystal engineering via donor-acceptor interactions. X-ray crystal structure and solid state reactivity of the 1:1 complex, 3,4-dimethoxycinnamic acid-2,4-dinitrocinnamic acid. *J Chem Soc, Chem Commun*, :45-46, 1983.
- [53] For example, C. B. Åakeroy, J. Desper and B. A. Helfrich. *CrystEngComm*, 6:19-24, 2004.
- [54] Ö. Almarsson and M. J. Zaworotko. Crystal Engineering of the Composition of Pharmaceutical Phases. Do Pharmaceutical Cocrystals Represent a New Path to Improved Medicines? *Chem Commun*, :1889 – 1896, 2004.
- [55] J. F. Remenar, S. L. Morissette, M. L. Peterson, B. Moulton, J. M. MacPhee, H. Guzmán and Ö. Almarsson. Crystal engineering of novel cocrystals of a triazole drug with 1,4-dicarboxylic acids. *J Am Chem Soc*, 125:8456-8457, 2003.
- [56] S. L. Childs, L. J. Chyall, J. T. Dunlap, V. N. Smolenskaya, B. C. Stahly and G. P. Stahly. Crystal Engineering Approach To Forming Cocrystals of Amine Hydrochlorides with Organic Acids. Molecular Complexes of Fluoxetine Hydrochloride with Benzoic, Succinic, and Fumaric Acids. *J Am Chem Soc*, 126:13335-13342, 2004.
- [57] S. Fleischman, L. Morales; B. Moulton, N. Rodríguez-Hornedo, R. Bailey Walsh and M. J. Zaworotko. Crystal engineering of the composition of pharmaceutical phases. *Chem Commun*, 186, 2003.
- [58] A. V. Trask, W. D. S. Motherwell and W. Jones. Solvent-drop Grinding: Green Polymorph Control of Cocrystallisation. *Chem Commun*, 890-891, 2004.
- [59] N. Variankaval, R. Wenslow, J. Murry, R. Hartman, R. Helmy, E. Kwong, S.-D. Clas, C. Dalton and I. Santos. Preparation and Solid-State Characterization of Nonstoichiometric Cocrystals of a Phosphodiesterase-IV Inhibitor and L-Tartaric Acid. *Cryst Growth Des*, 6:690-700 2006.
- [60] S. J. Hehm, B. Rodriguez-Spong and N. Rodriguez-Hornedo. Phase Solubility Diagrams of Cocrystal Are Explained by Solubility

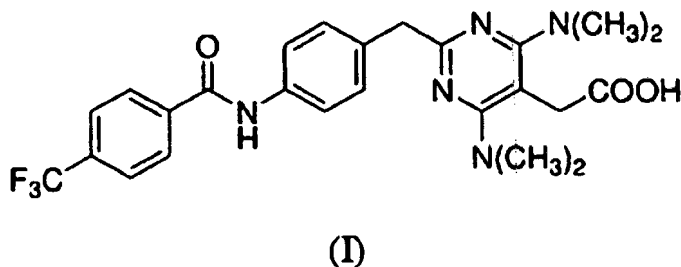
Product and Solution Complexation: Cryst
Growth Des, 6:592-600, 2006.

- [61] A. V. Trask, N. Shan, W. D. S. Motherwell, W. Jones, S. Feng, R. B. H. Tan and K. J. Carpenter. Selective Polymorph Transformation via Solvent-drop Grinding. Chem Commun., 880-882, 2005.

- [62] R. Storey, R. Docherty, P. Higginson, C. Dallman, C. Gilmore, G. Barr and W. Dong. Automation of Solid Form Screening Procedures in the Pharmaceutical Industry – How to Avoid the Bottlenecks. Crystallography Rev, 10(1):45-56, 2004.

REIVINDICACIONES

1. Una sal en forma cristalina, que comprende etilendiamina y ácido {4,6-bis(dimetilamino)-2-(4-(4-(trifluorometil)benzamido)benzil) pirimidin-5-il}acético de la Fórmula I:



que tiene un patrón de difracción de polvo de rayos X con un pico expresado en dos theta a aproximadamente 5.9°.

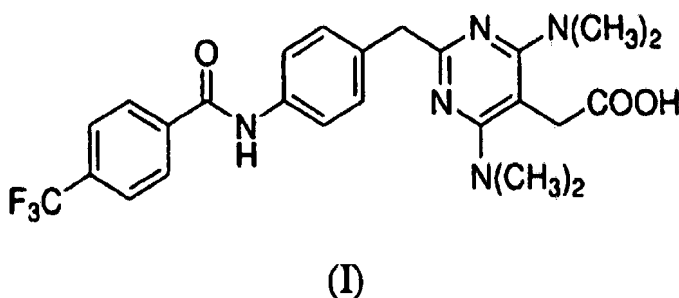
2. La sal de la reivindicación 1, que tiene un termograma de DSC con una endotermia a una temperatura pico de 123°C y una temperatura de inicio de 114°C.

3. La sal de la reivindicación 1 o 2, que tiene un termograma de DSC con una endotermia a una temperatura pico de 216°C y una temperatura de inicio de 212°C.

4. La sal de cualquiera de las reivindicaciones 1 a 3, que tiene que tiene una solubilidad acuosa de 6 mg/ml a 25°C.

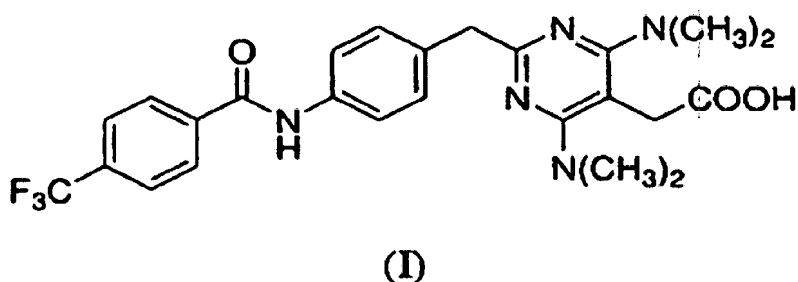
5. La sal de cualquiera de las reivindicaciones 1 a 4, que comprende de dos equivalentes molares del ácido de la Fórmula I y un equivalente molar de etilendiamina.

6. Una sal en forma cristalina, que comprende piperazina y ácido {4,6-bis(dimetilamino)-2-(4-(4-(trifluorometil)benzamido)benzil) pirimidin-5-il}acético de la Fórmula I:



que tiene un patrón de difracción de polvo de rayos X con uno o más picos expresados en dos theta a aproximadamente 10.7, 15.9, 22.3 y 24.0°.

7. La sal de la reivindicación 6, que tiene un patrón de difracción de polvo de rayos X con picos expresados en dos theta a aproximadamente 10.7, 15.9, 22.3 y 24.0°.
8. La sal de la reivindicación 6 ó 7, que comprende de dos equivalentes molares del ácido de la Fórmula I y un equivalente molar de piperazina.
9. La sal de cualquiera de las reivindicaciones 6 a 8, que tiene un termograma de DSC con una endotermia a una temperatura pico de 203°C y una temperatura de inicio de 98°C.
10. La sal de cualquiera de las reivindicaciones 6 a 9, que tiene un termograma de DSC con una endotermia a una temperatura pico de 223°C y una temperatura de inicio de 219°C.
11. La sal de cualquiera de las reivindicaciones 6 a 10, que tiene una solubilidad acuosa de 5 mg/ml a 25°C.
12. Una sal en forma cristalina, que comprende benzatina y ácido {4,6-bis(dimetilamino)-2-(4-(4-(trifluorometil)benzamido)bencil) pirimidín-5-il}acético de la Fórmula I:



que tiene un patrón de difracción de polvo de rayos X con uno o más picos expresados en dos theta a aproximadamente 8.0, 11.5, 16.0, 17.5, o 23.4°.

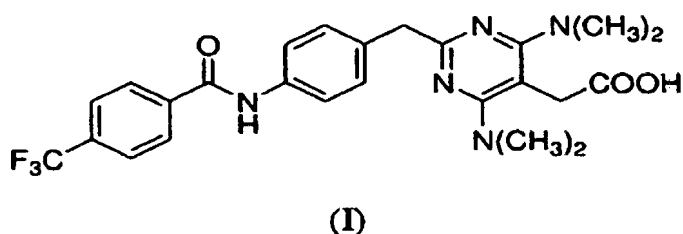
13. La sal de la reivindicación 12, que tiene un patrón de difracción de polvo de rayos X con picos expresados en dos theta a aproximadamente 8.0, 11.5, 16.0, 17.5, y 23.4°.

14. La sal de la reivindicación 12 ó 13, que comprende de dos equivalentes molares del ácido de la Fórmula I y un equivalente molar de benzatina.

15. La sal de cualquiera de las reivindicaciones 12 a 14, que tiene un termograma de DSC con una endotermia a una temperatura pico de 156°C y una temperatura de inicio de 154°C.

16. La sal de cualquiera de las reivindicaciones 12 a 15, que tiene una solubilidad acuosa de 0.08 mg/ml a 25°C.

17. Una sal en forma cristalina, que comprende colina y ácido {4,6-bis(dimetilamino)-2-(4-(4-(trifluorometil)benzamido)bencil) pirimidin-5-il}acético de la Fórmula I:



que tiene un patrón de difracción de polvo de rayos X con uno o más picos expresados en dos theta a aproximadamente 6.5, 19.6, 20.0, 21.9, o 26.1°.

18. La sal de la reivindicación 17, que tiene un patrón de difracción de polvo de rayos X con picos expresados en dos theta a aproximadamente 6.5, 19.6, 20.0, 21.9 y 26.1°.

19. La sal de la reivindicación 17 o 18, que comprende de un equivalente molar del ácido de la Fórmula I y un equivalente molar de colina.

20. La sal de cualquiera de las reivindicaciones 17 a 19, que tiene un termograma de DSC con una endotermia a una temperatura pico de 195°C y una temperatura de inicio de 193°C.

21. La sal de cualquiera de las reivindicaciones 17 a 20, que tiene una solubilidad acuosa de 24 mg/ml a 25°C.

22. Una composición farmacéutica que comprende la sal de cualquiera de las reivindicaciones 1 a 21, y un vehículo o excipiente farmacéuticamente aceptable.

23. La composición farmacéutica de la reivindicación 22, en donde la composición se formula para administración oral, nasal, bronquial o tópica.

24. La composición farmacéutica de la reivindicación 22 o 23, en donde la composición se formula como forma de dosificación individual.

