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Polymorphism and Migratory Chiral Resolution of the Free Base of Venlafaxine. A Remarkable Topotactical Solid State Transition from a Racemate to a Racemic Conglomerate

J. Th. H. van Eupen, W. W. J. Elffrink, R. Keltjens, and P. Bennema

Synthon BV, Microweg 22, 6545 CM, Nijmegen, The Netherlands

R. de Gelder and J. M. M. Smits

IMM, Chemical Crystallography, Radboud University Nijmegen, Toernooiveld 1, 6525 ED, Nijmegen, The Netherlands

E. R. H. van Eck and A. P. M. Kentgens

IMM, Solid State NMR, Radboud University Nijmegen, Toernooiveld 1, 6525 ED, Nijmegen The Netherlands

M. A. Deij, H. Meekes,* and E. Vlieg

IMM, Solid State Chemistry, Radboud University Nijmegen, Toernooiveld 1, 6525 ED, Nijmegen, The Netherlands

ABSTRACT: The unique behavior of the active pharmaceutical ingredient Venlafaxine free base, used as an antidepressant, with respect to polymorphism and chiral resolution is reported. Using several complementary techniques, three crystal structures of Venlafaxine were identified and isolated. All three structures are composed of virtually identical enantiomeric pure layers with different stacking modes. In the crystal structure with the highest melting point, the enantiomeric separation is complete, leading to a racemic conglomerate. The conglomerate can be grown from solution or via a solid–solid phase transition of the lowest melting racemic compound. Remarkably, the crystal shape is conserved during the transition. The corresponding chiral resolution is achieved via a local melting process, allowing for a long-range migration of the molecules between layers.

1. Introduction

Polymorphism, crystallization, and chiral resolution are phenomena that are of great industrial relevance. Here, we study a pharmaceutical compound, which combines these phenomena as a result of different stacking sequences of enantiopure layers of the compound in its various crystal structures.

The phenomenon of polymorphism, the ability of a substance to exist in two or more crystalline phases that differ in the arrangement and/or conformation of the molecules in the crystalline lattice, has important commercial and/or industrial implications in various fields.[1] The pharmaceutical industry and the photographic and imaging industry are two examples. Polymorphs consist of the same chemical compounds but have different crystal packings, and as a result, they can show different physical properties.[2] The occurrence of polymorphism is high, and McCrone stated that, in general, the number of polymorphic forms known for a given compound is proportional to the time and money spent in research on that compound.[3] Although the existence of different polymorphs of a certain compound is frequently observed, determining the conditions for their appearance usually is laborious. Most polymorphs are found as a result of serendipity rather than through systematic searches. The possibility to predict if and how many polymorphs exist of a compound is of great importance. Therefore, several computer programs with that aim have been developed, each of them with their specific advantages and limitations.[4,5] Most of these methods combine a random generation of crystal structures with an energy minimization step. An alternative way to predict the structures of possible polymorphs is the derived crystal packing (DCP) approach developed by Coquerel et al.[6] The DCP procedure comprises two steps: first, the extraction of periodic fragments (PF) from a known polymorphic form, and second, the generation of three-dimensional structures by the application of symmetry operators. New phases are then built as low energy structures by minimizing the energy using molecular mechanics software. Recently, the DCP method was successfully put into action to resolve the crystal structure of a metastable polymorph of Modafinil.[7]

Crystallization is often the final purification step for an active pharmaceutical ingredient (API). Until recently, most of the APIs were marketed as racemates. As a result of regulatory aspects, more and more effort is put into bringing enantiopure medicines to the market. In many cases this can be achieved by an enantioselective synthesis. Nevertheless, in an increasing number of cases, this would involve too many synthetic steps

* To whom correspondence should be addressed. E-mail: Hugo.Meekes@science.ru.nl.

for the process to be economically viable. Chiral separation by crystallization can be an alternative.

Crystalline racemates mainly belong to two classes. When both the enantiomers are present in equal amounts in the unit cell, the compound is called a racemic compound or a true racemate, which is the most common type. In 5–10% of the cases, racemic mixtures crystallize as racemic conglomerates, solids in which a single enantiomer is present in the unit cell. In these instances, a spontaneous segregation takes place. Often, one only considers a physical mixture of enantiopure crystals[8] as a racemic conglomerate. However, systems in which crystals are built up from macroscopic enantiopure layers, racemic twins, can also be considered as racemic conglomerates. This phenomenon was described by Gervais et al.[9] They investigated a compound which crystallizes as a racemic conglomerate but experienced a lot of trouble during separation by preferential crystallization. Isolated crystals, shaped as single crystals, showed no or almost no enantiomeric excess, a measure for the enantiomeric purity. This phenomenon was also observed by Green et al.[10] The initially considered (ordinary) macroscopic twinning along a growth direction was rejected as a result of dissolution experiments performed with the obtained crystals. Gervais et al.[9] explained the phenomenon as the repeated formation of epitaxial layers (macro-twinning) of crystals of the two enantiomers. Detailed study of the crystals revealed that the crystals were built up from thick alternate layers of *R* and *S* molecules, formed via an oscillating crystallization mechanism. An analogous case of spontaneous oscillating crystallization of *R* and *S* enantiomers was described by Potter et al.[11] and Berfeld et al.[12]

All these cases are a special case of epitaxial crystal growth. Usually epitaxy is observed as the oriented nucleation and growth of crystalline layers of a compound on a specific crystalline surface of another compound. This will often lead to large stresses and strains, as a result of the lattice mismatch. However, in the case of the epitaxy of enantiomers, a layer of, for example, *S* molecules grows epitaxially on top of a crystalline substrate of *R* molecules, without any significant lattice mismatch as a result of the enantiomeric relation between the two crystal structures. Note that the two layers do not need to be mirror images of each other. Nevertheless, the similarity between the two enantiomerically related structures can lead to a very low interfacial stress, resulting in a relatively low barrier for the epitaxial nucleation and growth. Epitaxial nucleation of strongly related crystalline phases was recently also found for polymorphs, that is, different crystal structures of the same compound.[13,14] In this case, the metastable polymorph of a hormone was found to grow epitaxially on the stable polymorph. In later studies, it was even found that the reverse process was also possible.[15] Both polymorphic forms consisted of almost identical layered structures. For the metastable polymorph, with spacegroup *P*1, only one conformer of the molecule was present, while the stable polymorph, with spacegroup *P*2₁, consisted of four successive layers having a different conformer of the molecule. Such a situation can be considered as a special case not easily covered by the DCP approach, as it involves different conformers of the molecule. Furthermore, in this case, the small structural difference between the layers resulted in a small barrier for epitaxial nucleation. The major difference with enantiomeric epitaxial crystallization, however, is the fact that the polymorphic epitaxial layers of the metastable polymorph were able to transform via a solution mediated mechanism to the stable polymorph structure. Giovannini et al.[16] described a transition of a true racemate to a racemic conglomerate. A racemic
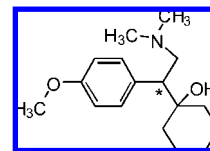
dihydrate of Zopiclone is dehydrated forming a metastable anhydrate which, upon further heating, melts, and the racemic conglomerate phase crystallizes out. A solid–solid transition of a racemic conglomerate to a true racemate is described by Mercier et al.[17] Racemic conglomerate crystals of the compound [{H₃N(CH₂)₂SS(CH₂)₂NH₃}PbI₅]·H₃O) changed conformation of half of the disulfide moieties upon heating to 75 °C, leading to a true racemate. The transition back to the racemic conglomerate could also be accomplished. One has to note, however, that the chirality of "chiral disulfides" often results from configurationally chiral ligands linked to the chalcogenide atoms and that racemization occurs in solution as a result of the relatively low barrier of rotation of the S−S bond between both enantiomeric conformers.

Here, we study Venlafaxine free base, which shows both polymorphism as well as chiral resolution as a result of different stacking sequences of enantiopure layers of the compound. Venlafaxine is an antidepressant of the class of phenethylamines that inhibits the reuptake of serotonin, norepinephrine, and, to a lesser extent, dopamine. It is administrated orally as the hydrochloric acid salt and is marketed as a racemate. Both enantiomers are reported to be active. The common name for the compound Venlafaxine is the following: 1-[2-(dimethylamino)-1-(4-methoxyphenyl)-ethyl]cyclohexanol. The molecular structure of the free base is given in Figure 1. Two polymorphs of the free base were known, and a previously performed extensive study of the thermal behavior of the free base yielded one new crystal structure.[18] It turned out that an enantiotropic[19] relation exists between all three crystal structures, implying that below a certain transition temperature one form is stable and above that temperature another one is stable. Form I is stable below 40 °C, form II is stable between 40 and 50 °C, and a third form is stable above 50 °C. The present study of the structural relation between the three crystal structures shows that forms I and II are true racemates built up of sequences of enantiopure *R* and *S* bilayers and enantiopure double bilayers, respectively, and, therefore, true polymorphs, while form III consists of alternate macroscopically thick layers of *R* and *S* molecules. Therefore, form III is an epitaxial racemic conglomerate and not a real polymorph in the strict sense of the definition. Furthermore, we investigate the metastable solid–solid transition (peritectoid) between the racemic compound form I and the racemic conglomerate, form III. Remarkably, this transition is topotactical, as the shape of the crystal does not change during this racemate to racemic conglomerate transformation.

2. Experimental Details

Venlafaxine with a purity of 99.9%, according to high pressure liquid chromatography (HPLC), was supplied by Synthon B.V. The solvents (pa) were purchased from Aldrich and used without further purification. Crystals of the three polymorphs were obtained by cooling a solution of Venlafaxine in heptane saturated at reflux temperature. The various forms were obtained by quickly cooling the clear solution to a temperature for which the desired form is stable[18] and isolating the formed crystals at that temperature. When isolating single crystals of

Figure 1. Molecular structure of Venlafaxine. The * symbol denotes the chiral carbon atom.

forms II and III, a layer of the low melting polymorph(s) is easily deposited on the crystal. To avoid the formation of these layers, water of the right temperature was poured in the reaction flask to separate the hot heptane solution from the crystals as fast as possible. After filtration of the suspension, the water attached to the crystals was removed by drying the crystals in a vacuum desiccator using phosphorus pentoxide to trap the water.

It was difficult to obtain suitable single crystals of the high melting form III for two reasons. First, crystallizing form III from solution often leads to very small crystals or an agglomerate of crystals. Second, form III is only stable above 50 °C, and because the solubility is very high at that temperature, 38 g of Venlafaxine "dissolves" in 10 g of heptane at 59 °C. Moreover, the liquid layer surrounding the isolated crystals cools quickly during isolation, and the resulting high supersaturation easily leads to the formation of a layer of polymorph I and/or polymorph II on top of the crystals of form III. Seeding a supersaturated solution above 50 °C with small crystals, isolated from a former experiment and washed to remove the layers of the other forms, resulted in the formation of large single crystals of form III.

Scanning electron microscope (SEM) photos were taken using a JEOL JSM 6330F field emission SEM. Optical rotations were measured with a Perkin-Elmer 343 polarimeter at 589 nm and 20 °C. The polarimeter was used with the micro-aperture setup.

For the single crystal X-ray diffraction measurements, single crystals were mounted in air on glass fibers. A course structure determination was performed at room temperature for each form. The final intensity data were collected at −65 °C; only a small contraction of the unit cell was observed. A Nonius Kappa CCD single-crystal diffractometer was used (φ and ω scan mode) using graphite monochromated Mo Kα radiation. Intensity data were corrected for Lorentz and polarization effects. For absorption correction, the Siemens Area Detector ABSorption correction program (SADABS)[20] was applied. All structures were solved by the program CRUNCH[21] and were refined with standard methods using SHELXL97[22] with anisotropic parameters for the nonhydrogen atoms. The hydrogens attached to the methyl and hydroxy groups were initially refined as rigid rotors to match maximum electron density in a difference Fourier map and were freely refined subsequently. All other hydrogens were initially placed at calculated positions and were also freely refined subsequently. For crystals of the enantiopure compound as well as crystals of form III (grown from solution or obtained by conversion of crystals of polymorph I after heating), the absolute structures could not be determined reliably by refinement of the Flack parameter in SHELXL97. However, based on the assignment of the chirality of Venlafaxine as in relation to the optical rotation as described by Yardley et al., we assigned the *R* conformation to the structure of the enantiopure compound.[23]

Cross polarization magic angle spinning (CPMAS) ^{13}C solid state NMR spectra[24,25] were acquired on a 400 MHz Chemagnetics Infinity spectrometer. For this analysis, 4 and 3.2 mm double resonant probes were used, tuned to 100.58 MHz for carbon and 399.95 MHz for protons. Measurements on polymorphs I, II, and the as-grown form III were done on the 4 mm probe with 8.0 kHz MAS. Here, variable amplitude cross polarization (VACP)[26] with a contact time of 2 ms was used with a radio frequency (RF) field strength of 64 kHz on protons and 56 kHz on carbons and a +1 to −1 linear ramp on protons. During acquisition, the protons were decoupled with a two pulse phase modulation (TPPM) pulse sequence[27] with a 110 kHz RF field, a pulse duration of 5.3 μs, and a phase modulation of 15 deg. Partially dissolved form III and enantiopure form III were measured with the 3.2 mm probe at a spinning speed of 12.5 kHz. VACP was used with a 62 kHz field with a +/−0.6 kHz ramp on ^{1}H and a 62 kHz field on ^{13}C. For proton decoupling, the continuous modulation (CM) scheme[28] was used with a modulation amplitude of 0.12 rad, a period of 8.4 μs, and an RF field strength of 110 kHz. Peak intensities were obtained by deconvolution of the spectra using a Voigt line shape. The data were processed using the MatNMR processing package which runs under Matlab.[29]

Molecular energies, lattice energies, and surface energies were calculated using the Cerius^2 modeling environment.[30] The crystal structures were minimized using the Dreiding force field with 3D-Ewald summation for the Coulomb contribution. Charges were determined using a restricted electrostatic potential (RESP) charge fitting scheme.[31] The Van der Waals interactions were calculated using a spline function with on- and off-distances at 12 and 13 Å, respectively. To calculate the lattice energy, the energy of individual molecules with the bulk

Table 1. Crystal Data of Forms I, II, III (Grown from Solution), III (after Transformation), and One of the Pure Enantiomers of Venlafaxine Free Base[a]

	space group	*a* (Å)	*b* (Å)	*c* (Å)	α	β	γ	Z	*V* (Å^3)
form I	P2$_1$/n	8.32	8.82	21.61	90	92.22	90	4	1586.4
form II	P2$_1$/c	8.84	8.27	43.75	90	90.97	90	8	3198.6
form III (grown)	P2$_1$2$_1$2$_1$	8.22	8.86	22.27	90	90	90	4	1622.7
form III (transformed)	P2$_1$2$_1$2$_1$	8.23	8.87	22.33	90	90	90	4	1630.5
pure enantiomer	P2$_1$2$_1$2$_1$	8.15	8.80	22.30	90	90	90	4	1599.3

^{a} All structures were measured at −65 °C.

conformation was calculated and subtracted from the energy found for the crystal structures. For the calculations of surface energies, the crystal structure was cleaved. Using the same settings as for the bulk crystals, only now with a 2D-Ewald summation, the surface energies were calculated by subtracting the lattice energy of the full crystal from that of the cleaved semi-infinite crystals without relaxation.

3. Results and Discussion

3.1. X-ray Single Crystal Structure Determination. The crystal structures were determined using single crystal X-ray diffraction. Polymorphs I and II crystallize in the monoclinic spacegroups P2$_1$/n and P2$_1$/c, respectively. Note that the latter spacegroup represents an alternative setting of the former one. Form III crystallizes in an orthorhombic structure, spacegroup P2$_1$2$_1$2$_1$. For an overview of the crystal data of the three polymorphs, see Table 1.

The crystal structure determinations of forms I, II, and III showed that all stuctures are built up from similar enantiopure layers parallel to the (001) surface. The crystal structure of polymorph I is in agreement with the previously published structure[32] (Cambridge Crystal Database reference code OCALAG). In the case of polymorph I, alternating layers of *R* and *S* molecules make up the structure; for polymorph II, alternating bilayers of *R* and *S* molecules are present; and for form III, only layers of one enantiomer make up the complete crystal packing. Polymorphs I and II are true racemates because *R* and *S* molecules are stoichiometrically, ratio (1:1), present in the unit cell. For the unit cells of forms I, II, and III, see Figure 2.

The crystallographic relationship between molecules in the enantiopure layers of forms I, II, and III differ slightly. In form I, the molecules in the enantiopure (mono)layers are related by translational symmetry in the *a*- or *b*-direction or by a 2-fold screw axis in the *b*-direction. In form II, the molecules in the enantiopure layers are related by translational symmetry in the *a*- and *b*-direction or by a pseudo-2-fold screw axis in the *a*-direction. In form III, the molecules in the layers are related in the same way as in form I. The consequence of this is that the molecular arrangement in the *a*-direction of form I is similar to the arrangement in the *b*-direction of form II and similar to the *a*-direction of form III. And, vice versa, the molecular arrangement in the *b*-direction of form I is similar to the arrangement in the *a*-direction of form II and similar to the *b*-direction of form III. This relationship is also visible from the unit-cell parameters of the three forms. The crystallographic relationship between layers consisting of different enantiomers is in both forms I and II, the inversion symmetry. The relation between layers of the same enantiomer that make up bilayers in the structures of forms II and III is a 2-fold screw axis, for form II in the *b*-direction and for form III in the *a*-direction. This means that inverting an enantiopure layer must yield a
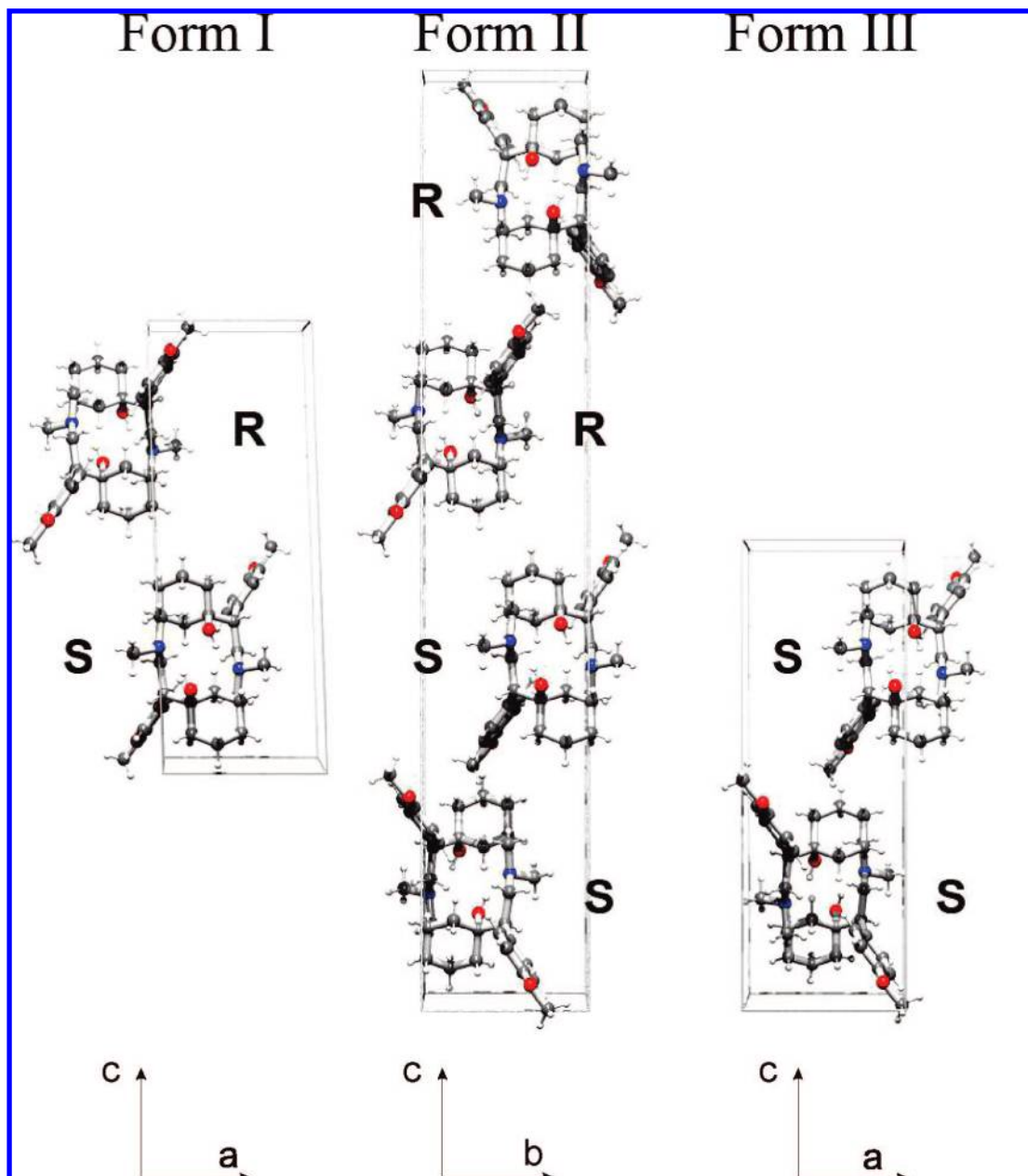
Figure 2. Projections of the unit cells of forms I, II, and III of Venlafaxine. For forms I and III, the *a* and *c* axes and, for form II, the *b* and *c* axes are indicated.

resulting layer that is structurally very similar to a layer obtained by applying a 2-fold screw axis (in the proper direction), at least with respect to the structure of the interface between two enantiopure layers. This structural resemblance is visible in Figure 3 in which an overlay is shown of *R* and *S* layers that have a similar contact to other enantiopure layers. Clearly, the overall structure of the interfaces is very similar although the layers are made up of different enantiomers. The crystal structure of form III pointed to the presence of only one enantiomer, although we used the same racemic starting material as for the crystallizations of polymorphs I and II. This indicates the formation of a racemic conglomerate, as will be discussed below.

3.2. Solid State NMR Characterization. Using ^{13}C solid state NMR all three forms were investigated. Figure 4 shows the spectrum of a microcrystalline powder of a pure enantiomer grown from a heptane solution. These crystals have the same structure as form III. The assignment of the carbon spectrum

was done based on chemical shifts and dipolar dephasing experiments (not shown here).[33] Peaks stemming from carbon 6 and 7a/b are somewhat broader due to the residual dipolar coupling to ^{14}N. Stacking effects are clearly present in the spectrum as carbon atoms that would be equivalent in solution now have different chemical shifts (up to 10 ppm for carbon 10a/b). Figure 5 displays the ^{13}C spectra of the microcrystalline powders which were grown from a racemic solution. Polymorphs I and II are shown in Figure 5a and b, while the spectrum of the as-grown form III and the partially dissolved form III are displayed in parts c and d. Clear differences can be seen between the different forms. One can observe three sets of chemical shifts; those shared between polymorphs I and II, those shared by forms II and III, and those shared by all three forms. Combining this with the crystal structures, it is straightforward to conclude that the chemical shifts shared between polymorph I and II are associated with the stacking of 2 different enantiomer layers, i.e. the *RS* interface between an *R* bilayer
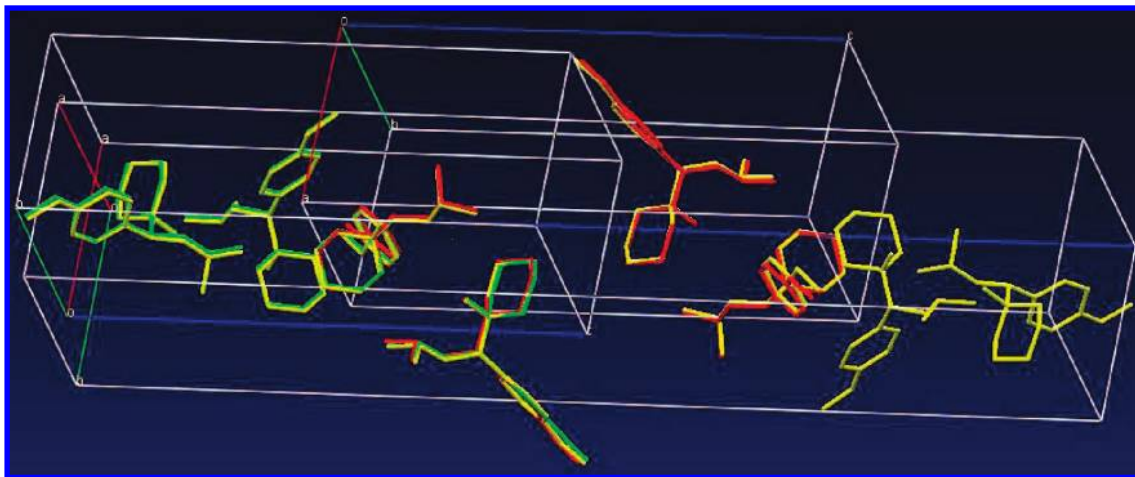
**Figure 3.** Overlay of the unit cells of polymorph I (green), polymorph II (yellow), and form III (red).
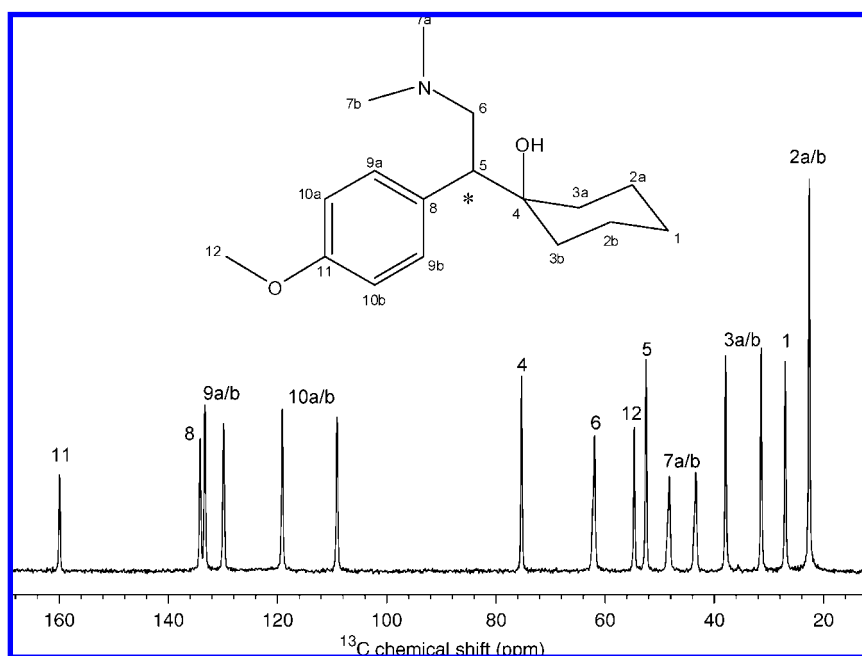
Figure 4. ^{13}C CPMAS NMR spectrum of Venlafaxine in form III, grown from an enantiopure solution. The numbers on top of the peaks correspond to the carbon atom labels in the molecular structure.

and an *S* bilayer, while those shared between II and III are unique for the stacking of like enantiomer layers, i.e. an *R* bilayer stacked on another *R* bilayer (or an *S* bilayer on another *S* bilayer). The difference in chemical shift between an unlike interface (*RS*) and the like interface (either *RR* or *SS*) are most noticeable for peaks stemming from carbons 1 and 12 and a little less noticable for the other 6-ring carbons (2a/b, 3a/b, and 4). From the crystal structure (see Figure 2), these are actually the carbon atoms that are lying at the interface of the bilayers. Quantification of ^{13}C CPMAS spectra has to be done with care and is usually not straightforward. In this case, however, no differences in CP dynamics between the different forms could be observed. For polymorph II, we know that half of the peaks originate from the like interface and the other half from the unlike interface. Inspection of the spectrum of form II shows that intensities of, for instance, the two peaks of carbon 12 appear in a 1:1 ratio. Hence, quantification of the relative amounts of the layer types based on the VACP measurements was possible.

3.3. Dissolution Experiments of Grown Crystals. The X-ray data of form III of Venlafaxine pointed toward the formation of a racemic conglomerate, with single crystals composed of enantiopure fragments. Furthermore, both the X-ray crystallography and the NMR results indicate that the structure of form III grown from the racemic solution is identical to the one grown from an enantiopure solution.

A possible explanation of the observation is that somehow during the process an enantiomeric resolution took place and separate crystals of *R* and *S* were formed. To examine this possibility, several large single crystals were isolated and the optical rotation of the single crystals was measured separately. In all cases, the optical rotation was negligible. From this result, it was concluded that (almost) equal amounts of both enantiomers were present in the crystals.

Therefore, during low supersaturation conditions the single crystals have to grow via the successive formation of enantiopure fragments. A mechanism, which accounts for the formation of
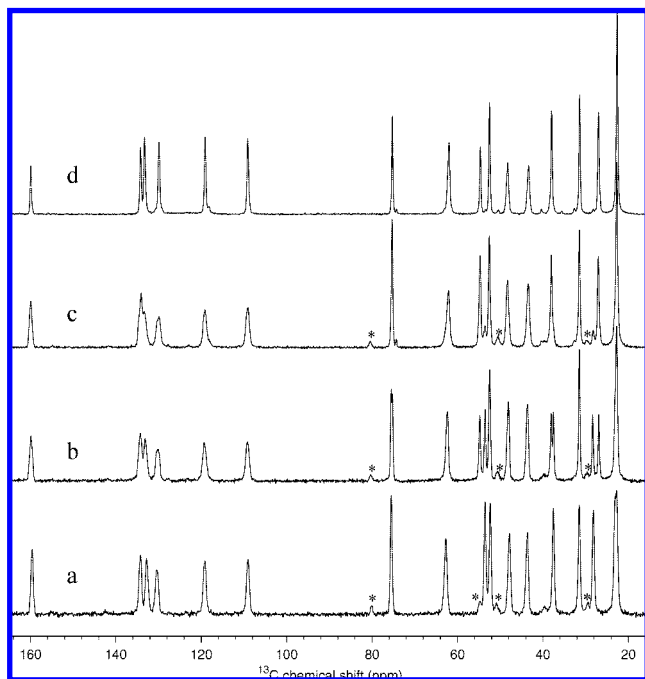
Figure 5. ^{13}C CPMAS NMR spectra of the different forms of Venlafaxine, grown from a racemic solution: (a) form I, (b) form II, (c) form III, as-grown, and (d) form III, rinsed. Chemical shifts are listed in Table 2. Spinning sidebands are indicated with an asterisk. MAS speeds were 8 kHz in parts a−c and 12.5 kHz in part d.

Table 2. ^{13}C **Chemical Shifts for the Different Forms**[a]

carbon atom	form I δ (ppm)	form II δ (ppm)	form III δ (ppm)
2a/b	22.7	22.7	22.7
		22.7	22.7
	23.2	23.2	
1	28.2	28.3	
		26.9	27.0
3a/b	31.5	31.5	31.4
	37.6	37.5	
		38.0	38.0
7a/b	43.6	43.6	43.4
	47.8	48.0	48.3
5	52.3	52.4	52.5
12	53.5	53.5	
		54.7	54.7
6	62.7	62.3	62.0
4		75.2	75.3
	75.5	75.5	
10a/b	109.1	109.1	109.2
	119.1	119.3	119.2
9a/b	130.4	130.1	129.9
	132.8	133.1	133.5
8	134.3	134.2	134.1
11	159.6	159.9	156.0

[a] Carbon atoms are numbered according to Figure 4.

single crystals consisting of macroscopic layers of enantiopure fragments, via oscillating crystallization, was already described in the literature.[9,11] During the growth of a face, only one enantiomer is inserted and, as a result of that, the local supersaturation of the other enantiomer increases until a certain threshold value. Then, nucleation of the other enantiomer on top of the already existing enantiopure fragment results in the growth of a new enantiopure fragment of the opposite handedness. Gervais et al. were able to overcome the problem of the formation of crystals by slowly stirring the solution. In that case, the diffusion rate is large enough to keep the supersaturation of the other enantiomer below the threshold of 2D nucleation. Instead of trying to avoid this oscillating growth behavior, we study the composition of the as-grown crystals in more detail.

To further investigate the composition of the single crystals, selective dissolution experiments were performed as proposed by Toyokura et al.[34] which offer a simple method to study the enantiomeric composition of the single crystals of form III. A crystal of form III which was grown from a heptane solution containing racemic Venlafaxine was added to a saturated solution of one of the pure enantiomers in heptane at room temperature. Assuming ideal solubility, mainly fragments containing the *R* enantiomer should dissolve in a solution saturated with the *S* enantiomer at a temperature for which form III crystals are metastable. The dissolution process was followed in situ using optical microscopy. After several hours, the crystals were isolated and analyzed using scanning electron microscopy (SEM). In Figure 6, the left image gives an overview of a crystal before etching. A striped pattern is visible on top of the crystal, suggesting a fragmented composition of the crystal present before etching. This pattern is shown enlarged in the inset and can be compared with the SEM image of a crystal after etching, shown on the right in Figure 6.

Some crystals were left in the solution saturated with the *S* enantiomer for 16 h. The optical rotation of the isolated

fragments showed an enantiomeric excess of more than 80%. This result confirms that mainly fragments containing *R* enantiomers dissolve. From a comparison of the SEM images, it is obvious that intermediate layers of the crystal, with opposite handedness, dissolved during the dissolution experiment. Moreover, the images confirm that the as-grown single crystals of form III are composed of enantiopure fragments with a thickness of 10–50 μm. These fragments contain one of the enantiomers in a large excess.

From the NMR data, one can also determine the average layer thickness through the relative ratio of the peaks unique for the unlike (*RS*) interface and the peaks unique for the like interface (*RR* and *SS*). The as-grown form III (Figure 5c) shows some low intensity peaks that are characteristic for the RS interface. On the basis of peak intensities, one arrives at an average like layer thickness between 8 and 10 interfaces (comparable to 4–5 stacked unit cells of form III). This seems rather thin, compared with the layer thickness as seen in the SEM images. However, as was mentioned before, the isolation of single crystals of form III caused some difficulties, the result of the high solubility of Venlafaxine in heptane. Crystals isolated from the solution are covered with a layer of liquid with a high concentration of Venlafaxine. A layer of polymorph I and/or II can crystallize on the surface of the crystal of form III before this liquid layer can be removed. Forms I and II both contain *RS* interfaces, and this would explain the existence of the low intensity peaks in Figure 5c. To examine this idea, single crystals of form III were added to a flask with heptane and left there for some time to dissolve the outer layer, the part of the crystal where *RS* interfaces are likely to be present. After isolation and drying of the crystals, the spectrum in Figure 5d was recorded. The intensities of the *RS* interfacial peaks are much reduced, leading to an average enantiopure layer thickness of 40 layers (equivalent to 20 unit cells of form III stacked along *c*). This is still much thinner than the 10–50 μm (which would be 5000−25 000 unit cells) which the SEM images suggested. It should be noted, however, that the layer thickness is an average over the entire crystal; enantiopure layers could be separated by thin layers of form I and/or II, or single layers of one enantiomer can be interspersed within a thick layer of the other enantiomer. These thin layers of what would essentially be a single unit cell of
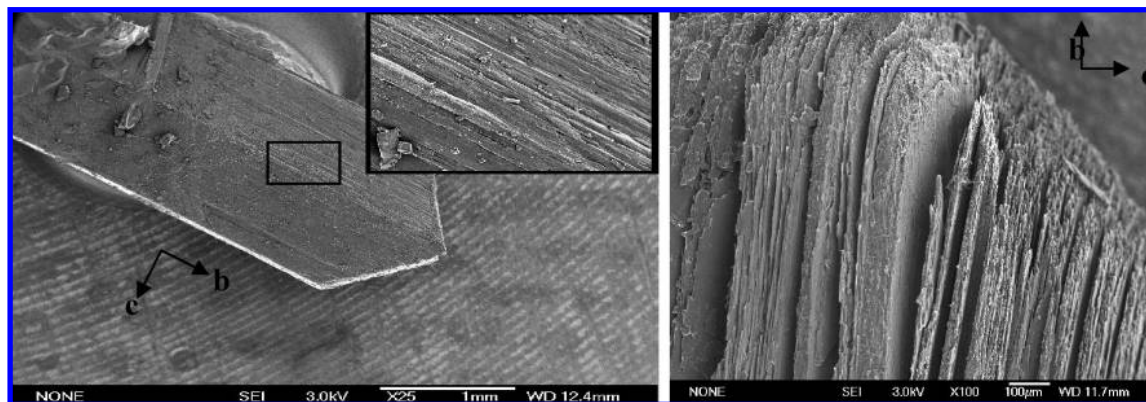
Figure 6. SEM photographs of single crystals of form III before (left; scale bar = 1 mm) and after (right; scale bar =0.1 mm) a dissolution experiment in a heptane solution saturated with one of the enantiomers. The insert shows a part of the crystal surface with the same magnification as for the image on the right. The orientation of the *b* and *c* axes is indicated.
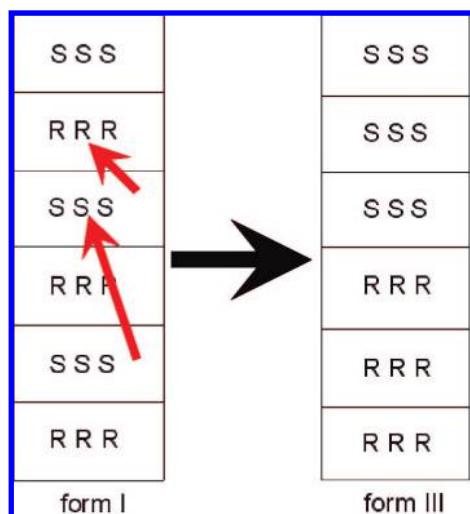
Figure 7. Annealing of a crystal of polymorph I at 75 °C for 6 days leading to its transformation to form III. The *R* and *S* enantiomers need to migrate over surprisingly large distances during this process.

form I would not show up in the X-ray diffraction, nor in the etching experiments.

3.4. Solid–Solid Transition of Polymorph I to III. In a related paper,[18] the solid–solid transition of form I to III was followed in time using differential scanning calorimetry (DSC), X-ray powder diffraction (XRPD), and solid state NMR. Especially the XRPD and NMR measurements showed a gradual transformation between the polymorphs. To realize such a transformation in the solid state, complete layers of *R* and/or *S* molecules have to be transferred, a process that on first sight seems very unlikely; see Figure 7.

For NMR, XRPD, and DSC, finely powdered material was used. Taking into account the large surface area and the acquired stress resulting from grinding, the solid–solid transition is imaginable. The question then arises, whether this transition could also be established in a single crystal. A comparable experiment was performed using a single crystal of polymorph I (dimensions approximately 2 mm in all directions). With single crystal X-ray diffraction, the crystal structure was confirmed. The single crystal was subsequently transferred to a calorimeter and slowly heated to 74 °C which is 4 °C below the melting temperature of form III (that is, the eutectic temperature of the racemic conglomerate), kept at that temperature for 6 days, and then cooled to room temperature. According to single crystal

structure determination, the starting material, polymorph I, was converted to form III; remarkably, the shape of the crystal had not changed.

To study the layered structure of this crystal in more detail, an annealed crystal of polymorph I was etched in a similar way as for crystals of form III by adding it to a saturated solution of one of the pure enantiomers. SEM photos were taken from the isolated crystals after the etch experiment; see Figure 8. Although the solution might lead to a reconstruction of the layered structures, it is clearly visible from the figure that the layers are built up in a mosaic fashion, contrary to the layered fragments of the etched crystals of form III as grown from solution; see Figure 6.

The only way to transform form I into III is by the migration of molecules, inversion of the chiral center is impossible. Furthermore, diffusion of complete layers in the solid state is also not very likely. To account for this phenomenon it is suggested that diffusion has to occur via a kind of molten phase. A possible explanation is local melting as a result of crystal defects, impurities and/or stress several degrees below the melting point of the bulk solid. During the annealing, a thin layer melts, and after nucleation, an *R* or *S* domain starts to grow. As in the experiment of Gervais et al.,[9] after some time, the supersaturation of the other enantiomer has reached the threshold for 2D nucleation and, after nucleation, an enantiopure domain starts to grow epitaxially on top of the underlying layer. For the present case, it is probable that the premelting process starts at different spots in the crystal simultaneously. At some of these places, *R* starts to nucleate, and at others, *S* will start to nucleate. In this way, no enantiopure layer, as for the crystals of form III grown from solution, but a mosaically composed layer is formed for the annealed crystal. The molten fronts move through the crystal resulting in a crystal composed of enantiopure domains. The thickness of the domains is of the same order as for the layers of the grown crystals of form III.

The difference between the volumes of the unit cells of the grown (1623 Å³) and the transformed form III (1630 Å³) confirm the view resulting from the above-described experiment. During the transformation of polymorph I to form III, the packing efficiency of the formed mosaical crystal fragments differed slightly from that of grown crystals of form III, resulting in a slightly larger unit cell. This is also validated by the temperature factors of the determined crystal structures. For the transformed form III, they are slightly larger.

3.5. Molecular Modeling. Looking at the remarkable solid–solid transition, one can ask what is the driving force for
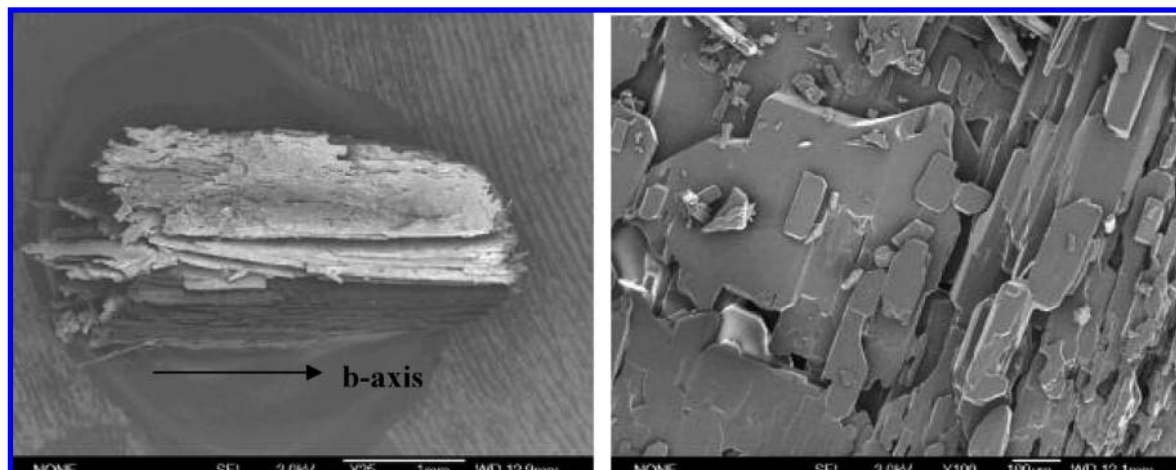
Figure 8. SEM photographs of annealed (6 days, 74 °C) crystals of polymorph I after a dissolution experiment in a heptane solution saturated in one of the enantiomers: (left) scale bar = 1 mm, magnification 25×; (right) scale bar =0.1 mm, magnification 100×, The orientation of the *b*-axis is indicated.

Table 3. Calculated Lattice and Interface Energies for Forms I, II, and III

	form I	form II	form III
crystal [kcal/unit cell]	67.67	161.49	61.93
molecule 1 [kcal/mol]	44.4	48.44	43.27
molecule 2		47.64	
lattice energy [kcal/mol]	−27.49	−27.85	−27.78
$R-R$ surface		178.19	77.97
$R-R$ excess energy		16.70	16.04
$R-S$ surface	82.73	177.34	
$S-S$ excess energy	15.03	15.85	

this racemate-to-conglomerate transition. As the structures of the individual layers for all three forms are virtually the same, the interactions between molecules inside a layer can be considered equal. For the interlayer interactions, it can be concluded that, at lower temperatures, for which form I is stable, $R-S$ interfaces are favored, whereas at higher temperatures (form III), $R-R$ (and $S-S$) interfaces are dominant. Form II can be considered as an intermediate phase as 50% of its interfaces are $R-S$ and the remaining 50% are $R-R$ and $S-S$.

To get an estimate of the energies involved in the interface, energies were determined using molecular mechanics calculations of the lattice energies of a bulk crystal and a cleaved one. The results of these calculations are summarized in Table 3. The lattice energies are the same within 0.4 kcal/mol, thus within the error margin (roughly 2 kcal/mol) of the calculations. The difference in surface energy between forms I and II is for the $R-S$ layers 0.8 kcal/mol, which may indicate a small enthalpic favor for form I and is in accordance with the fact that form I is formed at low temperatures. The difference in $R-R$ surface energy for forms II and III, however, is 0.7 kcal/mol, which hints at a small enthalpic favor for form III forming over form II, at lower temperatures, which is in contrast with the observations.

Entropic contributions usually dominate the polymorphic stability more than the enthalpic contributions.[19] Assuming that the transitions between the enantiotropically related forms are mainly entropy driven, in other words assuming that the enthalpies and entropies of the polymorphs differ but are only weakly dependent on temperature, the order of enthalpies is $H_I < H_{II} < H_{III}$ and the order of entropies follows $S_I < S_{II} < S_{III}$. The entropy has two contributions: a conformational part and a vibrational part. The conformational part is the same for all the three forms, whereas the vibrational part of the entropy will

differ. Limiting our attention again to the interlayer interactions and form III having only interactions between R and R (or S and S) layers, this leads to the conclusion that there is more vibrational freedom in these interactions as compared to interactions between R and S layers.

4. Conclusion

Three different forms of the antidepressant Venlafaxine were identified and their crystal structures determined. The crystals of the three forms are composed of enantiopure layers. Form I consists of alternating bilayers of R and S enantiomers, while form II consists of two R bilayers alternating with two S bilayers. Single crystal X-ray diffraction experiments combined with dissolution experiments showed that crystals of form III are racemic conglomerates, composed of a stacking of relatively thick layers of R and S enantiomers. Therefore in Venlafaxine, a unique combination of phenomena, polymorphism, and chiral separation is encountered in a single compound. The overall structure of the interfaces between the bilayers in the three polymorphs is very similar; the sequence of the bilayers, however, differs considerably for the different forms. From ^{13}C solid state NMR experiments, it emerged that each type of layer interface (RS and RR/SS) has a unique spectral signature. From these experiments, the average (enantiopure) layer thickness for form III was calculated. Although this led to much thinner layers compared with the layer thickness found from SEM images, showing that etching of the crystals in the presence of one of the enantiomers resulted in lamellae of 10–50 μm thickness, the results could be compared taking into account that the etching experiment will not reveal the thinnest lamellae present. Molecular modeling studies show that the differences in lattice enthalpy between the three structures is of the order of 1 kcal/mol, implying that the transitions between the three forms are entropy driven. Furthermore, it was shown that a single crystal of Venlafaxine form I readily undergoes a topotactical solid–solid phase transition to form III as the habit of the crystal did not change during this transformation. The experiments indicate that local melting is the underlying mechanism for this remarkable transformation as the long-range migration of molecules in the solid state needed to accomplish this transformation seems

highly unlikely. Further investigations to gain insight into the dynamics of this process are underway.

Supporting Information Available: Crystallographic data of all crystal structures in CIF format. This material is available free of charge via the Internet at http://pubs.acs.org.

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