Solid-State Analysis of Polymorphic, Isomorphic, and Solvated Forms of Dirithromycin

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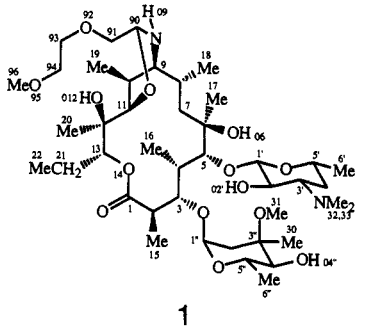
Abstract: Dirithromycin, a semisynthetic macrolide antibiotic, crystallizes in two anhydrous polymorphic forms, an amorphous form and at least nine stoichiometric solvate forms. Six of the known solvates are isomorphic, having nearly identical X-ray powder diffraction (XRPD) patterns. Differences are observed in the CP/MAS ^{13}C solid-state NMR spectra, which show resonances associated with the incorporated solvent molecules. Variable-temperature CP/MAS ^{13}C solid-state NMR spectra of the isomorphic solvates relate *N,N*-dimethylamine peak coalescence temperatures with steric hindrance of included solvent molecules. The crystal structures of the thermodynamically stable anhydrous polymorphic form, a 1-propanol solvate, and a cyclohexane trisolvate form are reported and compared to that of the previously reported acetonitrile trihydrate crystal form. The crystal packing, hydrogen-bonding networks, and molecular conformations are related to the spectroscopic properties of the various crystal forms and an amorphous form. The FTIR lactone carbonyl stretching frequency and the ^{13}C solid-state NMR chemical shift of the lactone carbon resonance are related to the presence or absence of hydrogen bonding to this group. This study shows the usefulness of these combined spectroscopic techniques for the study of solids with structures that have not been determined. Furthermore, the study demonstrates the wide range of crystallographic forms in which macrolide antibiotics exist.

Introduction

Dirithromycin (**1**), the 9-*N*-11-*O*-oxazine derivative of erythromycin [9*S*(*R*)-9-deoxo-11-deoxy-9,11-[imino[2-(2-methoxyethoxy)ethylidene]oxy]erythromycin], is an important member



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of the macrolide class of orally active antibiotics.[1-3] The structural features of erythromycin are a 14-membered lactone ring having two sugar moieties attached by a glycosidic bond, a cladinose sugar moiety (denoted by double-prime numbers) attached at the 3-position, and a desosamine sugar moiety (denoted by single-prime numbers) attached at the 5-position. The structure of a physically unstable, disordered acetonitrile trihydrate crystal form has been reported[4] as well as a closely related derivative of erythromycin, V-T 108.[5] The existence of other crystal forms of dirithromycin was first indicated by X-ray powder diffraction (XRPD) studies of samples recrystallized from different solvents. Subsequent research showed the existence of two crystalline, nonsolvated crystal forms, nine solvated crystal forms, and a form which was amorphous by XRPD. The studies herein were carried out, since the polymorphic system of dirithromycin permits examination of the effect of solid-state environment on the molecular properties of the macrolide and since relatively few structures of erythromycin derivatives have been determined.[5-7]

Solid-state NMR is a powerful technique for studying solids and the molecular mobility of solids.[8-11] Molecular mobility has been related to a number of the chemical and physical properties of solids, with chemical stability being a property of particular importance to pharmaceuticals.[12-15] Dirithromycin offers an excellent opportunity to use solid-state NMR for studies of molecular mobility because the molecule can be studied in various crystalline and noncrystalline environments and in the presence and the absence of solvent of crystallization. The solid-state NMR results reported in this paper show that the coalescence temperature of the *N,N*-dimethylamino group of the desosamine moiety depends upon the crystalline conformation and the solvent present in the crystal lattice. Furthermore, this paper shows that solid-state NMR is perhaps the most definitive technique for

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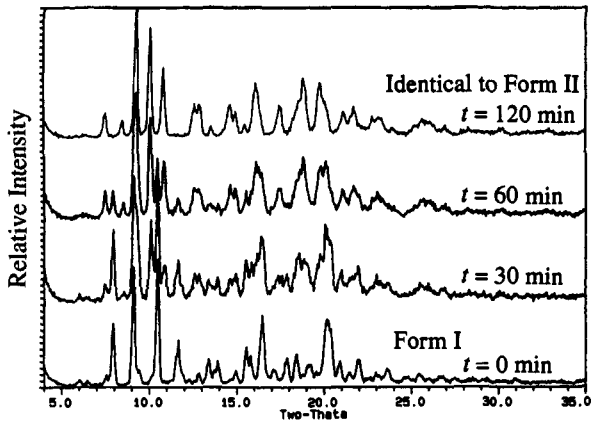

Figure 1. X-ray powder diffraction patterns of the isothermal phase transition of form I to form II at 110 °C.

characterization of amorphous materials related to erythromycin. In combination with other solid-state spectroscopic techniques, key information on the structure, material properties, and molecular mobility of solids can be obtained. This knowledge is particularly critical for pharmaceuticals, since the physical form often determines the rate of dissolution, bioavailability, and stability.[16]

Results and Discussion

X-ray powder diffraction studies provided the first indication of polymorphism in dirithromycin. Repeated recrystallizations from polar organic solvents gave mixtures of polymorphic forms I and II resulting from the drying process. Differential thermal analysis (DTA) and variable-temperature XRPD were used to determine that form I converted to form II at elevated temperatures (Figure 1). During the search for a solvent system which would provide pure form II crystals directly from solution, it was found that crystals isolated from acetone and 2-propanol gave XRPD patterns which were nearly identical but differed from those of forms I and II. It was postulated that the guest solvent molecule was incorporated in a cavity of a similar crystal lattice. Further recrystallizations were conducted in which the solvent molecule's size was varied to determine the approximate volume of the cavity. It was found that when the solvent reached the size of 2-methyl-2-propanol, there was a dramatic change in the XRPD pattern. Since this system appeared to be one of the best examples of polymorphic and isomorphic solvates and the dirithromycin molecule in the acetonitrile trihydrate crystal form was shown to adopt the less common "folded in" macrolide conformation, attempts were then made to grow single crystals of the various crystalline forms.[4,17,18]

X-ray Crystallography. Form II, the 1-propanol solvate, and the cyclohexane trisolvate were determined to be monoclinic $P2_1$ (No. 4), whereas the previously reported acetonitrile trihydrate[4] and the similar compound V-T 108[5] were determined to be orthorhombic $P2_12_12$ (No. 18). The crystal data of the three structures reported herein are provided in Table 1.

Figure 2 provides a visual comparison of dirithromycin conformations in the four different crystal forms and shows that, in each structure, the aglycone ring moiety adopts the well characterized "folded in" conformation.[17,18] This is in contrast to the structure of VT-108, in which the aglycon adopts the "folded out" conformation. This change in conformation is due to the configuration about C90—in dirythromycin it is *R* whereas in

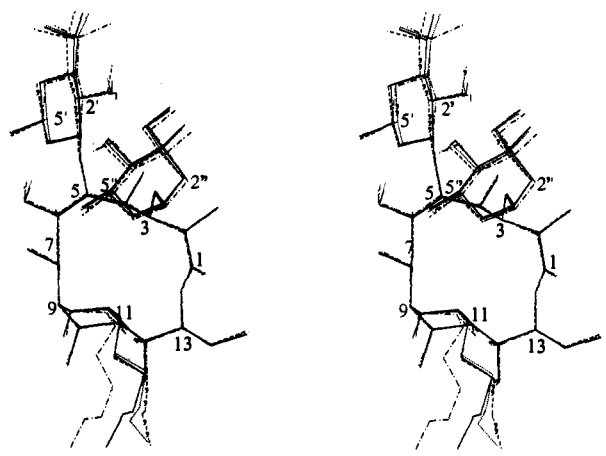

Figure 2. Superposition of dirithromycin molecular conformations in cyclohexane trisolvate (—), form II (- - -), acetonitrile-trihydrate (– – –), and 1-propanol solvate (– · – · –) crystals.

VT-180 it is *S*.[19] Unit cell packing diagrams of the crystal structures reported herin (form II, the 1-propanol solvate, and the cyclohexane trisolvate crystals) as well as the acetonitrile trihydrate form previously reported are shown in Figure 3. A comparison of the torsion angles illustrates the close similarity of the four dirithromycin structures with the greatest degree of conformational variation to be associated with the *N,N*-dimethylamino group of the desosamine moiety and the polyether side chain.

Hydrogen Bonding. The intermolecular hydrogen-bonding network of form II, the cyclohexane trisolvate, and the acetonitrile trihydrate crystals consist of a series of hydrogen bonds between the lactone carbonyl oxygen (O1) of one molecule and the cladinose hydroxyl group (–O4″–H04″) of a second molecule, forming a continuous strand of molecules arranged in a helical fashion. The O1···O4″ distances in the three structures are 2.80(1), 2.82(1), and 2.79(1) Å, respectively. In the acetonitrile trihydrate structure, the hydrogen bonding of the solvent molecules was not reported by the authors because of disorder.[4]

In the 1-propanol solvate structure, the intermolecular hydrogen-bonding network differed from the previous two solvate structures in that lactone was not hydrogen bonded. The solvent molecule was hydrogen bonded to the desosamine nitrogen (2.79(2) Å O101···N3′). The thermal parameters, bond lengths, and bond angles indicate that the solvent molecule is disordered relative to the rest of the structure. The positions of the solvent molecule's carbon atoms and its oxygen atom however are reported in the structure because of their relevance to future discussion. The intramolecular hydrogen bond distances of the structures are reported in Table 2.

Isomorphic structures in which the solvent is purely electron donating, such as acetone or 2-butanone solvates, would not possess the hydrogen bonding of the solvent molecule exhibited in the 1-propanol solvate structure. Because of this, it is inferred that other forces dictate the overall molecular packing in the isomorphic structures. There are numerous hydrogen bond donor and acceptor sites in the dirithromycin molecule, however, few of which participate in intermolecular hydrogen bonds. Instead, many of them are not satisfied or deviate significantly from linearity in the four structures which have been determined. This may be due to conformational restrictions of the macrolide which limit the molecule's ability to simultaneously form multiple intermolecular hydrogen-bonding contacts. The various structures show that the hydroxyl groups are located within the interior of the molecule while the hydrophobic groups are positioned

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Table 1. Crystal Data, Data Collection, and Refinement for Form II, 1-Propanol Solvate, and Cyclohexane Trisolvate

	form II	1-propanol solvate	cyclohexane trisolvate
exp formula	$C_{42}H_{79}N_2O_{14}$	$C_{42}H_{79}N_2O_{14}\cdot C_3H_8O$	$C_{42}H_{79}N_2O_{14}\cdot 3C_6H_{12}$
formula wt	835.10	895.19	1087.58
crystal dim (mm)	0.49 × 0.35 × 0.22	0.39 × 0.26 × 0.20	0.30 × 0.29 × 0.20
crystal color	colorless	colorless	colorless
crystal shape	needle	needle	rhomboid
crystal system	monoclinic	monoclinic	monoclinic
space group	$P2_1$ (No. 4)	$P2_1$ (No. 4)	$P2_1$ (No. 4)
a (Å)	12.254(2)	14.563(3)	16.024(4)
b (Å)	13.123(1)	11.777(5)	12.932(2)
c (Å)	14.507(1)	14.785(3)	16.309(2)
β (deg)	103.61(1)	94.15(2)	101.34(1)
V (Å^3)	2267.4(8)	2529(2)	3313(2)
Z	2	2	2
δ_{calc} (g·cm^{-3})	1.223	1.175	1.090
temp (K)	293	293	293
radiation (λ)	Mo Kα (0.710 73 Å)	Mo Kα (0.710 73 Å)	Cu Kα (1.541 84 Å)
monochrometer	graphite	graphite	none
linear abs coef (cm^{-1})	0.85	0.81	5.77
h	−14 to 14	0 to 17	0 to 17
k	0 to 15	0 to 13	0 to 13
l	0 to 17	−17 to 17	−17 to 17
2θ range (deg)	4.00–50.00	4.00–50.00	4.00–112.00
scan width (deg)	0.43 + 0.35 $tan\,\theta$	0.78 + 0.35 $tan\,\theta$	0.71 + 0.15 $tan\,\theta$
F_{000}	912.0	980.0	1200.0
p-factor (weighting)	0.040	0.040	0.040
no. of unique data collected	4188	4685	4540
data with $I > 3.0\sigma(I)$	3077	2171	4165
no. of variables	541	419	646
largest shift/esd in final cycle	0.42	0.01	0.54
R	0.042	0.070	0.082
R_w	0.051	0.077	0.109
goodness of fit	1.308	1.676	3.937

outward. This conformational arrangement may account for the pharmaceutical's exceptionally low water solubility despite the abundance of polar substituents associated with the sugar residues and the macrolide itself.[20]

X-Ray Powder Diffraction. X-ray powder diffraction patterns corresponding to each form of dirithromycin are provided in the supplementary material. The amorphous form of dirithromycin shows two very broad XRPD maxima of low intensity, thus indicating that there is a very low degree of order. The XRPD patterns of polymorphic forms I and II show major differences throughout their diffraction patterns. The possibility that form I is a mixture of form II and a solvate is discounted because there is no loss of volatiles as shown by thermal gravimetric analysis. Additionally, the X-ray powder diffraction pattern of form I is distinctly different from that of form II and the solvated crystal forms from which it is made. Furthermore, variable-temperature XRPD studies clearly show the disappearance of peaks associated with form I crystals and the appearance of form II peaks when form I is held isothermally at 110 °C (Figure 1). XRPD patterns of the isomorphic crystal forms of dirithromycin are very similar and in many cases indistinguishable within the experimental error of the technique. The diffraction patterns of the nonisomorphic crystal forms of dirithromycin are dramatically different, with each form being readily distinguished.

Solid-State ^{13}C NMR Spectroscopy. Solid-state ^{13}C NMR peak assignments were made by comparison with the solution spectrum of dirithromycin in combination with interrupted decoupling experiments.[21] In each solution spectrum, the solvent molecule which was incorporated in the crystal lattice was identified by its ^{13}C resonances after the respective solvated crystal form was dissolved and analyzed in deuterated chloroform. This was particularly interesting for the unusual cyclohexane trisolvate. Four dirithromycin methine proton resonances at 5.22 (1H), 4.92 (1H), 4.80 (1H), and 4.61 (1H) ppm with the six cyclohexane methylene resonance at 1.39 (12H) ppm were integrated to confirm the stoichiometry of the solvate. An integration ratio of 4.0:35.2 was found, clearly indicating that the drug to solvent stoichiometry was 1:3.

Figure 4 shows the solid-state ^{13}C NMR spectra of the amorphous and two anhydrous forms of dirithromycin, with the amorphous form giving much broader resonances, resulting in the resolution of fewer peaks. In the solid-state NMR spectrum of polymorphic form I, there are two resolved peaks in the carbonyl region at 177.7 and 179.9 ppm. XRPD showed that this crystal form was not merely a mixture of forms but rather was a phase distinctly different from all others. Since the molecule has only one lactone functionality, it is likely that there are two independent molecules in the asymmetric unit of the crystal lattice. The chemical shift data associated with the 1-propanol solvate and form II crystals, in which the lactone carbonyl is not hydrogen bonded in the former and is hydrogen bonded in the latter, can be used as a reference for the effect of hydrogen bonding on the chemical shift of the different crystal forms of dirithromycin. We propose this chemical shift difference of the two resonances of form I to be due to hydrogen bonding: one independent molecule having a hydrogen-bonded lactone carbonyl whereas the other is not hydrogen bonded or has a much weaker hydrogen bond. Similarly, the resonance of the lactone carbonyl of the amorphous form indicates it is free of hydrogen bonding. The direction and magnitude of the shift observed in dirithromycin are comparable to those observed in five crystal forms of prednisolone *tert*-butylacetate.[15]

Figure 5 demonstrates the unique ability of solid-state NMR to distinguish between each of the isomorphic crystal forms of dirithromycin. There are significant differences throughout each of the spectra which permit identification. Resonances of the incorporated solvent molecule are particularly evident in the spectra of the ketone solvates.

The spectrum of the 2-methyl-2-propanol solvate form of dirithromycin (Figure 6) is dramatically different from those of

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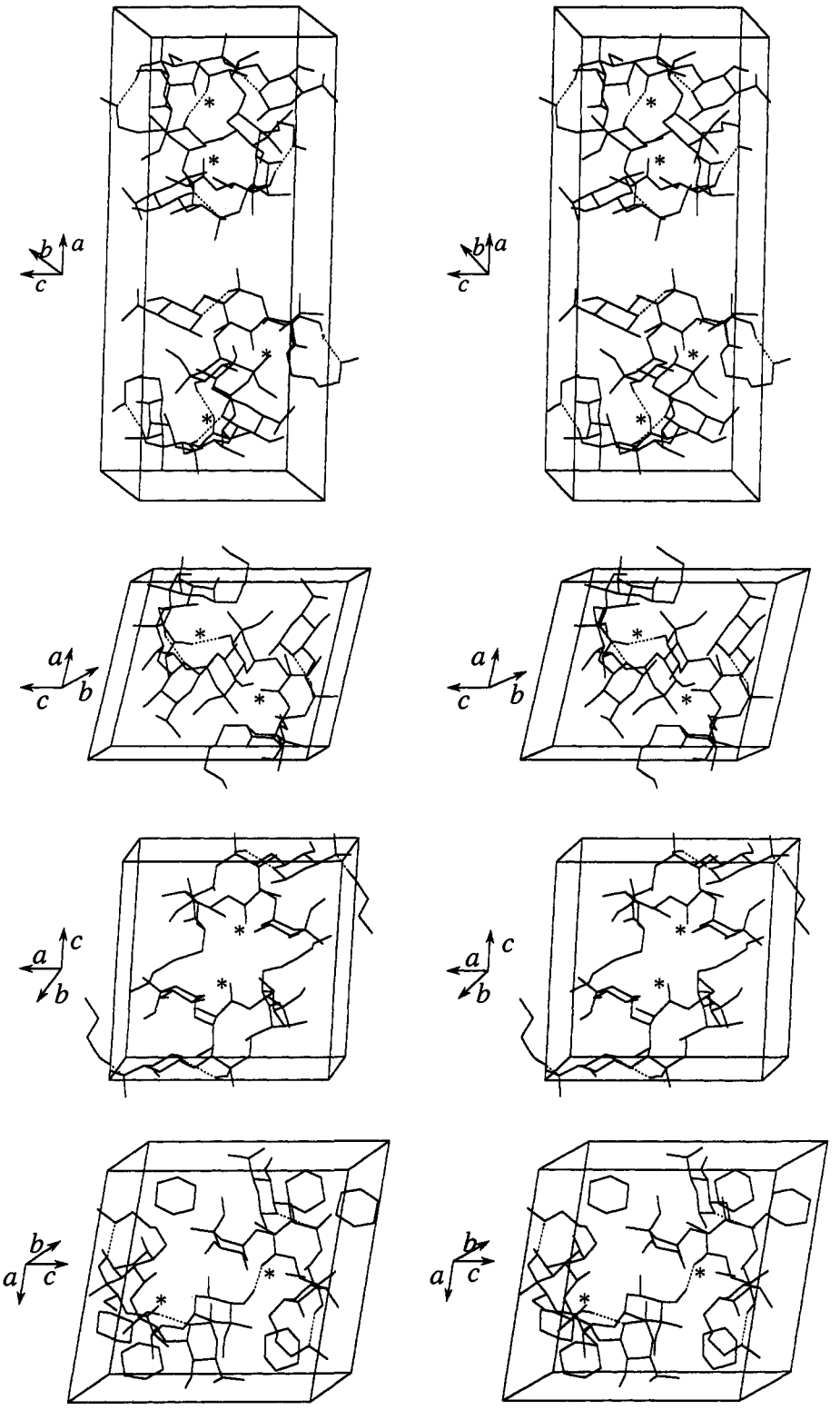

Figure 3. Stereoview of unit cell packing diagrams of acetonitrile trihydrate, form II, 1-propanol solvate, and cyclohexane trisolvate (top to bottom). Hydrogen bonds are indicated with dotted lines (those that involve molecules in other unit cells are not displayed). The carbonyl oxygens (O1) are indicated with asterisks.

all other forms; there is an upfield shift of the terminal methyl (C96) resonance of the polyether side chain, 55.6 ppm versus approximately 60 ppm in all other forms. This chemical shift difference may be attributed to *gauche* versus *trans* conformations of the polyether side chain.[22-24] In all four crystal structures, three reported herein and the acetonitrile trihydrate, the side chain adopts a conformation in which this methyl group is *trans* with respect to the γ-carbon.

Interrupted decoupling experiments at room temperature showed a relatively intense *N,N*-dimethylamine carbon resonance in most of the solid forms, but this was of reduced intensity in the 1-butanol solvate and was completely absent in the 2-methyl-

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Table 2. Geometry of the Inter- and Intramolecular Hydrogen Bonds in Dirithromycin Structures

X–H···Y	X···Y (Å)	H···Y (Å)	angle (deg)	equivalent position[a]
		Form II		
O6–H06···O5′	3.01(1)	2.20(1)	151	x, y, z
O4″–H04″···O1	2.80(1)	2.01(1)	176	$1-x, {}^1/_2+y, 2-z$
		1-Propanol Solvate		
O101–H0101···N3′	2.79(2)	1.70(1)	175	x, y, z
O6–H06···O5′	2.93(1)	2.30(1)	143	x, y, z
		Cyclohexane Trisolvate		
O6–H06···O5′	2.95(1)	2.20(1)	171	x, y, z
O12–H012···O95	3.15(1)	2.37(1)	150	x, y, z
O4″–H04″···O1	2.82(1)	1.79(1)	156	$1-x, {}^1/_2+y, 2-z$

[a] The equivalent position refers to the hydrogen atom acceptor.

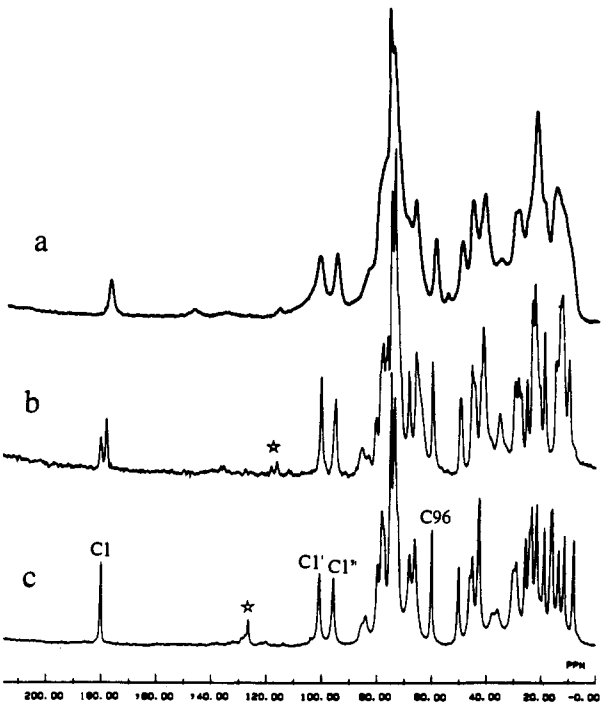

Figure 4. CP/MAS ^{13}C solid-state NMR spectra of anhydrous polymorphic forms: (a) amorphous; (b) less stable polymorphic form I (two carbonyl resonances due to two molecular conformations in the unit cell); (c) stable polymorphic form II. (Spinning side bands (star) and pertinent carbon atom resonances have been labeled.)

2-propanol solvate crystal form. Numerous solution NMR studies of trialkylamines have shown that there may be a combination of rotational and/or inversion processes which contribute to the observation of site-exchange effects.[25,26] In Figure 7, variable-temperature interrupted decoupling experiments on the 1-butanol solvate show two distinct carbon resonances at 46.3 and 34.3 ppm at 200 K which coalesce at approximately 218 K and are a single resonance at 40.9 ppm at 308 K. The coalescence temperatures for the 2-methyl-2-propanol solvate and the 1-propanol solvate crystal forms were approximately 298 and 200 K, respectively. The inversion and rotational process would require the breaking and formation of hydrogen bonds with the solvent. There is evidence that hydrogen bonds in solids are not necessarily static in nature.[27] In the four structures of dirithromycin, the 1-propanol solvate shows the amino nitrogen to be inverted with respect to those of the other three structures. The remainder of the desosamine ring and the macrolide itself are virtually superimposable. It is apparent that the protic solvent molecule provides an alternative energy minimum for the *N,N*-dimethylamino group conformation.

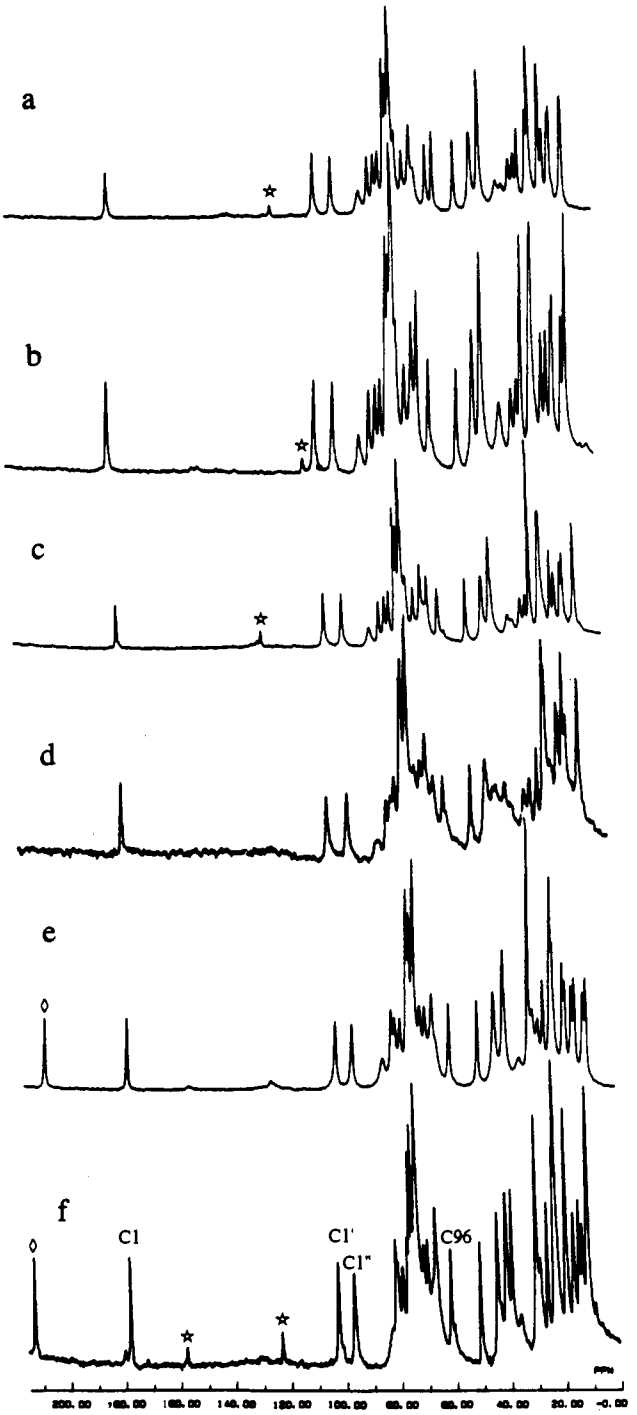

Figure 5. CP/MAS ^{13}C solid-state NMR spectra of isomorphic solvates: (a) ethanol solvate; (b) 1-propanol solvate; (c) 2-propanol solvate; (d) 1-butanol solvate; (e) acetone solvate; (f) 2-butanone solvate (significant differences are observed throughout the spectra despite similarities of crystallographic packing). (Spinning side bands (star), identified solvent resonances (diamond), and pertinent carbon atom resonances have been labeled.)

FTIR Spectroscopy. The FTIR spectra of the various crystal forms were very similar except for the O–H stretching frequencies, presumably reflecting differences in hydrogen bonding. In each of the spectra, a substantial amount of free O–H stretching is indicated. The lactone carbonyl region was most informative. X-ray crystallography has shown that the lactone functionality is hydrogen bonded in form II crystals but not in the 1-propanol

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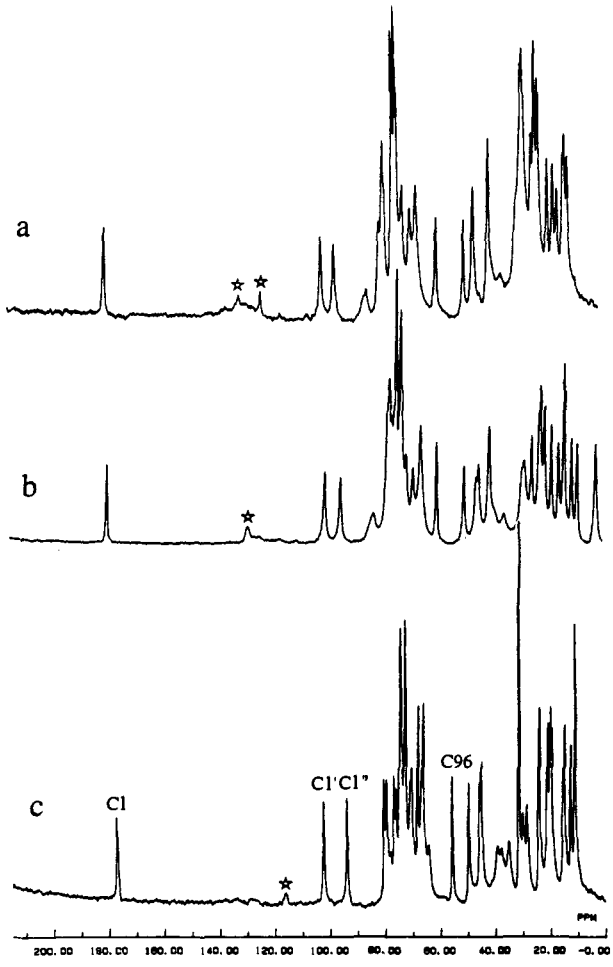

Figure 6. CP/MAS ^{13}C solid-state NMR spectra of nonisomorphic solvates: (a) cyclohexane trisolvate; (b) acetonitrile trihydrate; (c) 2-methyl-2-propanol solvate. The dramatic shift in the resonance of the C96 methoxy resonance of the polyether side chain is due to the "*γ-gauche*" effect. (Spinning side bands (star) and pertinent carbon atom resonances have been labeled.)

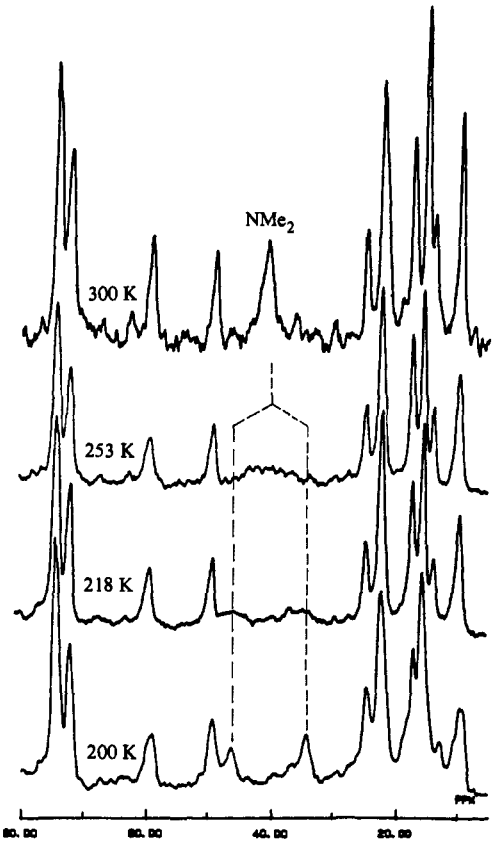

Figure 7. Variable-temperature interrupted decoupling ^{13}C solid-state NMR experiment of the 1-butanol solvate form indicating the occurrence of site exchange of the methyl resonances in the *N,N*-dimethylamino group. At high temperatures one resonance is observed; at lower temperatures slow exchange results in observation of the individual methyl group resonances.

solvate crystal form. The FTIR spectra are consistent with this observation. Thus, the lactone carbonyl stretching frequency of form II is shifted from that of the 1-propanol solvate crystal form (1712 and 1735 cm^{-1}, respectively). The direction and magnitude of the shift are consistent with other studies of hydrogen bonding in solids.[28] It is apparent that in the amorphous form of dirithromycin, the carbonyl is free of hydrogen bonding, since its vibrational frequency is the same as that of the 1-propanol solvate crystal form. Polymorphic form I crystals have stretching frequencies which are characteristic of both hydrogen-bonded and non-hydrogen-bonded lactone carbonyls. All of the isomorphic solvates have a carbonyl stretching frequency characteristic of the non-hydrogen-bonded lactone functionality (supplementary material). Interpretation of the acetone solvate and 2-butanone solvate spectra is complicated by the additional carbonyl of the incorporated solvent molecule. In the case of the nonisomorphic solvates, the carbonyl stretching frequencies of the cyclohexane trisolvate and acetonitrile trihydrate correspond to the hydrogen-bonded lactone carbonyl stretching frequency of form II. The lactone carbonyl stretching frequency of the 2-methyl-2-propanol solvate crystal form corresponds with that of the non-hydrogen-bonded 1-propanol solvate crystal form. Another interesting feature is that the crystal forms possessing a hydrogen-bonded lactone can be seen to have a shoulder on the peak which is of higher wavenumber (less hydrogen bonding character) when the sample is ground during preparation of its KBr pellet. It is thought that this may be due to disorder being introduced into the hydrogen-bonding network. Examination of spectra of single crystals of dirithromycin by FTIR microscopy confirmed this to be the case, with one very sharp absorption at 1711.7 cm^{-1}.

The presence of the acetonitrile solvent molecule is readily apparent in the spectrum of the acetonitrile trihydrate form of dirithromycin, with peaks at 2251.8 cm^{-1} and a smaller one at 2294.9 cm^{-1}. The presence of two stretching frequencies indicates that not all of the acetonitrile is hydrogen bonded in this crystal form and is attributed to disordered solvent molecules within the crystal lattice. This is consistent with the disorder observed by the authors in the structural report of this form.[4]

Thermogravimetric Analysis. Thermogravimetric analysis indicates that the loss of volatiles by the solvated crystal forms is stoichiometric and that they have discrete inflections at the onset temperature of solvent loss. The solvent loss observed for some of the crystal forms depends on the history of the sample after preparation; the ethanol solvate and the acetonitrile trihydrate crystal forms desolvate upon prolonged exposure to air.

Conclusion

Dirithromycin crystallizes in a number of different crystal forms. In this paper the X-ray crystallographic structures of form II, the 1-propanol solvate form, and the cyclohexane trisolvate form of the macrolide are reported, The molecular conformations of dirithromycin in these three crystal forms and the previously reported acetonitrile trihydrate form are very similar with the greatest degree of variability being in the polyether side chain. Significant differences in the intermolecular hydrogen-

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bonding networks were observed and are consistent with the ^{13}C solid-state NMR and FTIR spectra exhibiting distinct chemical shifts and absorption frequencies that are associated with hydrogen-bonded and non-hydrogen-bonded lactone functionalities. Our results show that FTIR is a powerful tool for studying hydrogen bonding in solids and provides important structural information on noncrystalline forms. ^{13}C Solid-state NMR provides an excellent advantage over other techniques in its ability to distinguish between different solvates of very similar crystallographic structure. Furthermore, variable-temperature ^{13}C solid-state NMR provides structural detail regarding temperature-dependent dynamics in the solid state. Further studies using variable-temperature ^{13}C solid-state NMR in combination with variable-temperature X-ray crystallography are planned to provide greater understanding about the dynamic processes in dirithromycin crystals.

Experimental Section

Materials. Dirithromycin was generously provided by Eli Lilly and Company.

Preparation of Crystal Forms. The polymorph designated form II was produced at room temperature by diffusion of 1-pentane into an ethyl acetate solution of dirithromycin or, alternatively, by water slurry of form I crystals at 50 °C for 30 min. Form I crystals were prepared by desolvation of the acetone solvate crystal form by vacuum drying at 40 °C overnight. The amorphous form of dirithromycin was obtained by freeze drying of the material from a methylene chloride solution The cyclohexane trisolvate was isolated by vapor diffusion of cyclohexane into a toluene solution of dirithromycin. Acetonitrile trihydrate crystals were isolated from a solution of the compound in water–acetonitrile after allowing slow evaporation of the solvent mixture. Dirithromycinethanol, 1-propanol, 2-propanol, acetone, 1-butanol, 2-butanone, and 2-methyl-2-propanol solvate crystals can each be isolated by slow evaporation of the solvent from the corresponding solutions.

Single Crystals. Polymorphic form II was obtained by diffusion of pentane into an ethyl acetate solution of dirithromycin over a period of 1 week. The 1-propanol solvate was grown by slow evaporation of a 1-propanol solution. The cyclohexane trisolvate was grown by vapor diffusion of cyclohexane into an ethyl acetate solution of dirithromycin over a period of approximately 2 weeks.

X-Ray Structure Determinations. All the crystal structures were determined and refined using similar procedures. Data were collected on an Enraf–Nonius CAD4 diffractometer. Three standard reflections were measured every 97 reflections; no crystal decay was detected. Lorentz and polarization corrections were applied to the data; no corrections were made for absorption. The structures were solved by direct methods using *SHELXS86*,[29] and the remaining atoms were located in succeeding difference Fourier synthesis. Only the hydrogen atoms bonded to heteroatoms were located and their positions and isotropic thermal parameters refined; the other hydrogens were located and added to the structure factor calculations but were not refined. The structures were refined in full-matrix least-squares using MolEN,[30] where the function minimized was $\sum w(||F_o|-|F_c||)^2$ and the weight w is defined per the Killean and Lawrence method[31] with terms of 0.020 and 1.0. Atomic scattering factors and the values for $\Delta f'$ and $\Delta f''$ were taken from *International Tables for X-ray Crystallography*.[32] Anomalous dispersion effects were included in F_c.[33] The final difference Fourier map showed no significant residual electron density, and the highest peak had a maximum ρ of 0.43 e/Å^3 with an estimated error based on a ΔF of 0.05.[34] Plots of $\sum w(||F_o|-|F_c||)^2$ versus $|F_o|$, reflection order in data collection, $\sin(\theta/\lambda)$, and various classes of indices showed no unusual trends.

X-ray Powder Diffraction. Samples were prepared by sieving a thin layer of sample onto a low-background sample holder. The Nicolet I2V diffractometer was operated in the step scan mode using a 2θ step size of 0.02° and a count time of 3 s. The instrument was equipped with a graphite diffracted beam monochromator and copper radiation source. The XRPD pattern was collected by measuring the scintillation response to Cu Kα radiation versus the 2θ value over a 2θ range of 4–35°. Variable-temperature XRPD was carried out using an Anton Paar TTK thermal accessory. The sample was held isothermally for variable lengths of time prior to data collection.

NMR Spectroscopy. CP/MAS ^{13}C solid-state NMR spectra were recorded at 50.19 MHz on a Chemagnetics M200 FT NMR spectrometer.[35] Approximately 250–300 mg of powdered sample was placed in a Kel-F rotor and spun at approximately 2.5–3.5 KHz. Free induction decays were defined typically by 8 K over a 3-KHz sweep width and were accumulated over 300–3000 transients with a recycle delay of 5 s for an acceptable signal-to-noise ratio. A proton decoupling field of 199.58 MHz and contact time of 2.00 ms were used. Chemical shifts were measured relative to a hexamethylbenzene (HMB) external standard with a methyl resonance at 17.36 ppm relative to tetramethylsilane.[36] Interrupted decoupling experiments were conducted using a 50-μs delay time prior to acquisition.[37] Variable-temperature CP/MAS ^{13}C solid-state NMR was conducted by monitoring the temperature using a thermocouple located in an exhaust gas port adjacent to the spinning sample. Calibration of the sample temperature was performed using the Curie-dependent chemical shift of samarium acetate tetrahydrate.[10]

Solution ^{13}C NMR spectra were obtained using Varian 500 and Varian 300 instruments. The spectra were acquired to identify the incorporated solvent molecules of dirithromycin. Each solvate was dissolved in deuterated chloroform at concentration of 50 mg/mL and its spectrum collected at 27.0 °C. Spectral widths of 33 and 20 KHz were used in the acquisition of 4000 scans at the ^{13}C observed frequencies of 125.697 and 75.429 MHz, respectively. The solution ^1H NMR spectrum of the cyclohexane trisolvate was obtained using a spectral width of 8 KHz in the accumulation of 128 scans at 499.843 MHz. The chemical shifts were referenced to an internal reference tetramethylsilane at 0.00 ppm.

Infrared Spectroscopy. A Nicolet 20SXC FTIR spectrometer with a DTGS detector was used to record the IR spectra. All spectra were from 128 coadded double-sided interferograms obtained at 2.0-cm^{-1} resolution and apodized with a Happ–Genzel function prior to transformation. Solid dirithromycin samples were gently ground with KBr and pressed into pellets. FTIR microscopy was performed using a Nicolet 60 SXB FTIR spectrometer. A Spectra Tech microscope was operated in the reflectance mode using a liquid nitrogen cooled, mercury–cadmium–telluride detector. The spectra resulted from the coaddition of 256–1024 single-sided interferograms obtained at 2.0-cm^{-1} resolution and again apodized using a Happ–Genzel function prior to transformation.

Thermal Analysis. Thermogravimetric analysis was performed to quantify the loss of volatiles over a range of 25–200 °C using a Dupont 951 thermogravimetric analyzer at a heating rate of 5 deg/min and a nitrogen flow rate of 5 mL/min. Differential thermal analysis was conducted using a Rigaku TG/DTA with a heating rate of 5 deg/min.

Form II. ^{13}C NMR (50 MHz, solid-state, 298 K, vs HMB external) δ 179.6, 100.2, 95.3, 84.8, 79.2, 77.8, 77.5, 76.8, 74.8, 73.6, 72.9, 71.7, 67.7, 65.8, 65.1, 59.6, 49.8, 45.6, 44.4, 42.3, 37.4, 35.4, 29.8, 29.1, 28.6, 25.5, 23.8, 23.2, 21.5, 21.5, 18.7, 16.1, 15.7, 13.3, 11.4, 7.9.

Acetonitrile Trihydrate. ^{13}C NMR (50 MHz, solid-state, 298 K, vs HMB external) δ 180.2, 101.2, 95.4, 83.3, 78.5, 77.8, 76.9, 75.4, 73.8, 73.1, 71.6, 69.1, 66.5, 65.5, 60.6, 50.5, 46.2, 45.2, 41.3, 39.2, 36.1, 29.4, 28.5, 25.8, 23.4, 22.7, 21.2, 18.7, 16.1, 14.0, 11.3, 9.3, 2.6.

Acetone Solvate. ^{13}C NMR (50 MHz, solid-state, 298 K, vs HMB external) δ 207.2, 177.1, 101.4, 95.3, 84.4, 81.2, 79.8, 77.9, 75.4, 74.5, 73.0, 70.7, 68.9, 67.2, 66.3, 64.9, 60.1, 49.8, 43.7, 40.2, 34.7, 31.3, 29.8, 27.9, 25.9, 23.2, 22.3, 20.4, 18.7, 17.6, 15.4, 14.5, 11.2, 10.2.

Form I. ^{13}C NMR (50 MHz, solid-state, 298 K, vs HMB external) δ 179.9, 177.7, 100.0, 94.8, 80.0, 77.9, 77.3, 76.0, 74.5, 73.2, 68.3, 65.5, 64.4, 59.5, 49.9, 49.0, 45.1, 44.3, 38.5, 35.0, 29.5, 28.4, 27.3, 25.1, 23.0, 22.1, 20.7, 18.7, 14.7, 14.3, 12.9, 12.2, 10.0.

2-Methyl-2-propanol Solvate. ^{13}C NMR (50 MHz, solid-state, 298 K, vs HMB external) δ 177.1, 102.3, 93.8, 80.2, 79.3, 76.6, 75.4, 74.5, 72.7, 70.8, 70.2, 68.1, 66.3, 64.3, 55.6, 49.6, 45.5, 45.2, 39.3, 37.8, 35.1, 31.4, 29.8, 28.4, 24.0, 20.9, 20.0, 19.5, 15.2, 14.8, 12.6, 11.0.

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1-Propanol Solvate, ^{13}C NMR (50 MHz, solid-state, 298 K, vs HMB external) δ 177.1, 101.8, 94.9, 84.9, 81.4, 79.2, 77.4, 75.4, 74.5, 73.1, 71.3, 68.4, 66.1, 64.4, 60.1, 49.7, 43.9, 41.4, 40.4, 36.7, 34.1, 29.9, 27.9, 26.5, 26.1, 22.6, 21.8, 18.5, 17.0, 15.1, 11.2, 10.1.

2-Propanol Solvate, ^{13}C NMR (50 MHz, solid-state, 298 K, vs HMB external) δ 177.1, 101.5, 95.0, 84.7, 81.3, 79.2, 77.8, 75.4, 74.6, 73.1, 71.4, 68.8, 66.1, 64.4, 63.8, 60.1, 49.8, 43.7, 41.0, 34.4, 32.9, 29.6, 27.9, 26.3, 22.6, 18.7, 17.4, 15.0, 14.3, 10.1.

Cyclohexane Trisolvate, ^{13}C NMR (50 MHz, solid-state, 298 K, vs HMB external) δ 179.7, 100.9, 96.2, 85.6, 84.2, 80.5, 79.9, 78.6, 75.1, 73.9, 71.5, 68.6, 66.5, 59.0, 49.0, 45.6, 40.1, 29.9, 28.0, 24.5, 23.4, 22.0, 18.6, 16.6, 15.0, 12.9, 12.4, 11.2.

2-Butanone Solvate, ^{13}C NMR (50 MHz, solid-state, 298 K, vs HMB external) δ 211.20, 177.1, 101.5, 95.1, 84.6, 81.3, 79.5, 77.7, 75.4, 74.5, 73.0, 71.2, 68.9, 66.2, 64.6, 63.9, 60.1, 57.8, 49.6, 43.7, 40.4, 38.3, 34.5, 29.5, 27.9, 26.0, 22.6, 18.6, 17.3, 15.1, 14.5, 10.7, 10.3.

1-Butanol Solvate, ^{13}C NMR (50 MHz, solid-state, 298 K, vs HMB external) δ 177.1, 102.4, 95.0, 83.7, 80.5, 79.1, 77.6, 75.2, 74.7, 72.9, 72.9, 70.0, 68.0, 66.1, 63.4, 59.8, 49.8, 44.2, 41.2, 40.3, 37.0, 34.5, 30.1, 28.0, 25.3, 22.8, 22.3, 19.7, 18.0, 15.8, 14.5, 10.3.

Ethanol Solvate, ^{13}C NMR (50 MHz, solid-state, 298 K, vs HMB external) δ 177.1, 103.0, 95.4, 84.4, 81.2, 79.5, 78.1, 75.8, 75.1, 73.3, 70.5, 68.5, 66.6, 63.9, 60.2, 50.1, 45.6, 41.0, 37.4, 35.1, 30.7, 28.5, 25.8, 23.3, 20.3, 18.4, 16.3, 15.1, 10.7.

Amorphous, ^{13}C NMR (50 MHz, solid-state, 298 K, vs HMB external) δ 177.0, 101.0, 94.9, 83.6, 78.0, 74.6, 73.3, 71.5, 69.0, 66.1, 58.9, 54.8, 49.3, 45.1, 42.5, 40.7, 35.1, 29.7, 28.4, 25.5, 21.6, 19.0, 14.5.

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Supplementary Material Available: The fractional coordinates of the three crystal structures reported herein (also submitted to the Cambridge Crystallographic Data Centre, University Chemical Laboratory, Lensfield Road, Cambridge CB2 1EW, U.K.), NMR vs FTIR lactone carbonyl correlation data for the various polymorphs, isomorphs, and solvates discussed, the gravimetric analysis of solvate stoichiometry, the X-ray powder diffraction data of the interplanar spacings and relative intensities of the various forms discussed, torsion angles of the three crystal structures, and an ORTEP drawing of form II (11 pages), This material is contained in many libraries on microfiche, immediately follows this article in the microfilm version of the journal, and can be ordered from the ACS; see any current masthead page for ordering information.