*Journal of Thermal Analysis and Calorimetry, Vol. 68 (2002) 453–465*

# SOLID STATE CHARACTERIZATIONS OF PHARMACEUTICAL HYDRATES

*D. Giron, Ch. Goldbronn, M. Mutz, S. Pfeffer, Ph. Piechon and Ph. Schwab*

Chemical and Analytical Research and Development, Novartis Pharma, Basel, Switzerland

## Abstract

Manufacturing processes may involve the presence of water in the crystallization of the drug substance or in manufacturing or in the composition of the drug product through excipients. Dehydration steps may occur in drying, milling, mixing and tabletting processes. Furthermore, drug substances and drug products are submitted to different temperatures and relative humidities, due to various climatic conditions giving rise to unexpected hydration or dehydration aging phenomena.Therefore the manufacture and the characterization of hydrates is part of the study of the physical properties of drug substances.

Several hydrates and even polymorphic forms thereof can be encountered. Upon dehydration crystal hydrates may retain more or less their original crystal structure, they can lose crystallinity and give an amorphous phase, they can transform to crystalline less hydrated forms or to crystalline anhydrous forms.

The proper understanding of the complex polyphasic system hydrates–polymorphs–amorphous state needs several analytical methods. The use of techniques such as DSC-TG, TG-MS, sorption-desorption isotherms, sub-ambient experiments, X-ray diffraction combined with temperature or moisture changes as well as crystal structure and crystal modelling in addition to solubilities and dissolution experiments make interpretation and quantitation easier as demonstrated with some typical examples.

**Keywords:** aging, DSC, freezable water, phase changes, phase transitions, pseudo-polymorphism, stability, sub-ambient DSC, TG, TG-MS, water activity

## Introduction

Manufacturing processes may involve the presence of water in the crystallization of the drug substance or in the manufacturing or in the composition of the drug product through excipients. New phases where water is a part of the crystal, called hydrates may be obtained with completely new properties in the solid state [1–8]. Dehydration steps may occur in drying, milling, mixing and tabletting processes. Some properties known to be altered by the association of solids with water, including rates of chemical degradation in the solid state, crystal growth, dissolution, dispersibility, wetting, powder flow, lubricity, compactibility, hardness. Furthermore drug substances and

*1418–2874/2002/ $ 5.00*

*Akadémiai Kiadó, Budapest*
*Kluwer Academic Publishers, Dordrecht*

drug products are submitted to different temperatures and relative humidities, due to various climatic conditions giving rise to unexpected hydration or dehydration aging phenomena. Water will be absorbed and desorbed with temperature and moisture changes. The crystallization in tablets [10, 11] of theophylline monohydrate, which has a lower dissolution rate as the two anhydrates [9] or the hydrate formation of excipients [12] such as lactose, sorbitol or magnesium stearate are among well known examples studied in the literature [3].

Several hydrates and even polymorphic forms thereof can be encountered. Upon dehydration crystal hydrates may retain more or less their original crystal structure, they can lose crystallinity and give an amorphous phase, they can transform to crystalline less hydrated forms or to crystalline anhydrous forms. In addition to physical changes, free water may react chemically [13]. Since the formation of new hydrated solid phases has the same impact as polymorphism for the bioavailability, toxicity, stability and processing as pure polymorphs, the manufacture and the characterization of hydrates is part of the study of the physical properties of drug substances according to the ICH decision tree 4 [14].

*Experiments were performed with the following instruments*

Perkin Elmer DSC-7 with robot system, Sub-ambient Perkin Elmer DSC-7, Perkin Elmer TGA-7, Mettler TA 850, Mettler TG-MS, X-ray diffractometer Scintag XDS 2000 with autosampler or with heating cell or with humidity cell for the X-ray diffraction experiments and DVS for the sorption-desorption curves. Crystal modelling was done using the Cerius software (MSI).

## Examples

*Phase diagrams considerations*

In crystal hydrates, the combination of intermolecular forces (hydrogen bonding) and crystal packing can produce very strong water-solid interactions. However, they are situations where hydration and dehydration of crystals occur quite easily at low temperature. The equilibrium behaviour of the anhydrous form of a drug substance with water depends upon the phase diagrams which are drug substance specific. Figure 1 shows two typical phase diagrams for a compound formation in a binary mixture. Figure 1a deals with the case of a hydrate which has a well defined melting point and Fig. 1b corresponds to an incongruent behaviour. Such phase diagrams are responsible for the observation of the eutectic point in the thermal analysis studies as it has been described in the case of Terpin [15].

By varying the temperature and the humidity different stable hydrates may be formed. When coming back to different conditions, a reconversion may be observed upon aging. An old example is emetine dihydrochloride for which the heptahydrate (15.0–19.0% water) and the pentahydrate (11.0–15.0% water) are described in the European Pharmacopea. In 1975 we analysed a reference declared as heptahydrate (loss on

drying 18.1%, water by Karl Fisher: 17.3%). 3 months later a conversion into the pentahydrate was suggested by the values of the loss on drying in a drying oven (14.7%), the thermogravimetry (14.8%) and the Karl Fischer determination of water: 14.8%.

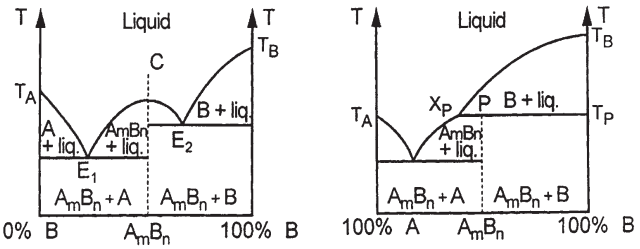


**Fig. 1** Phase diagrams of binary mixtures. 1a – compound with melting, 1b – incongruent behaviour

Figure 2 examplifies the DSC behaviour of the hydrates of two different drug substances with the dehydration without melting at very different temperature. For even less stable hydrates, the endotherm of dehydration may be so broad, that it can be difficult to detect it by only DSC and TG determinations. The ultimate demonstration of the hydrate formation is the single X-ray structure.

Since it is not always available, it has been suggested [16] to use sub-ambient DSC in order to know if the water contained in the sample is tightly bound or not. The melting peak of water allows to determine the freezable water.

The example given in the Fig. 3 deals with a crystalline drug substance for which a monohydrate structure was suggested. No freezable water was found in the sample, even after exposition at 92% RH. The drug substance was slurried with water in order to maximise the uptake of water and the sample analyzed by TG and sub-ambient DSC. The TG value of the sample was 33.8%. The freezable water cal-

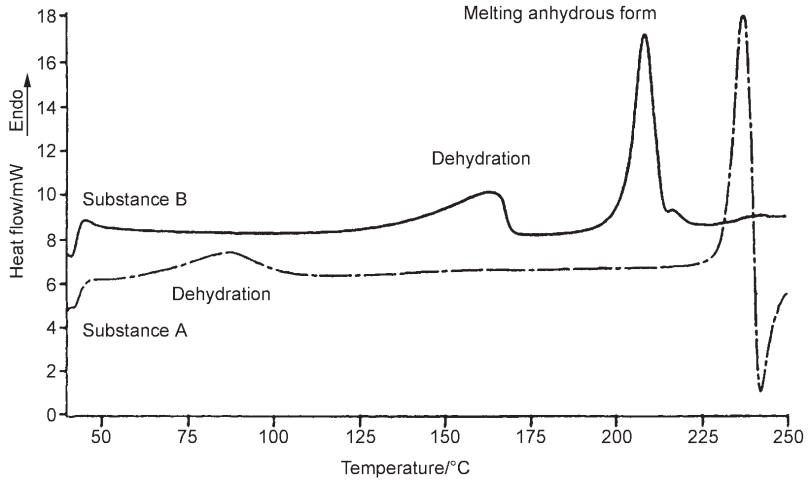


**Fig. 2** DSC scans of two hydrated forms with different energies resulting in different temperatures for the dehydration endotherms

culated 31.4%. Therefore the amount of hydrated solid was 68.6% and the amount of bound water calculated 3.5%, which fit exactly with the theoretical amount of water for a monohydrate.

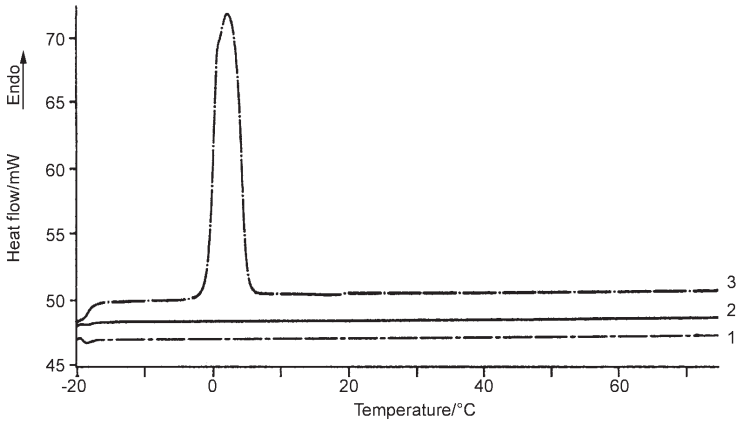


**Fig. 3** Sub-ambient DSC and TG experiments for the study of bound water
1 – Drug substance; 2 – drug substance stored under 92% RH; 3 – suspension of the drug substance with water. Melting peak of free water in the suspension

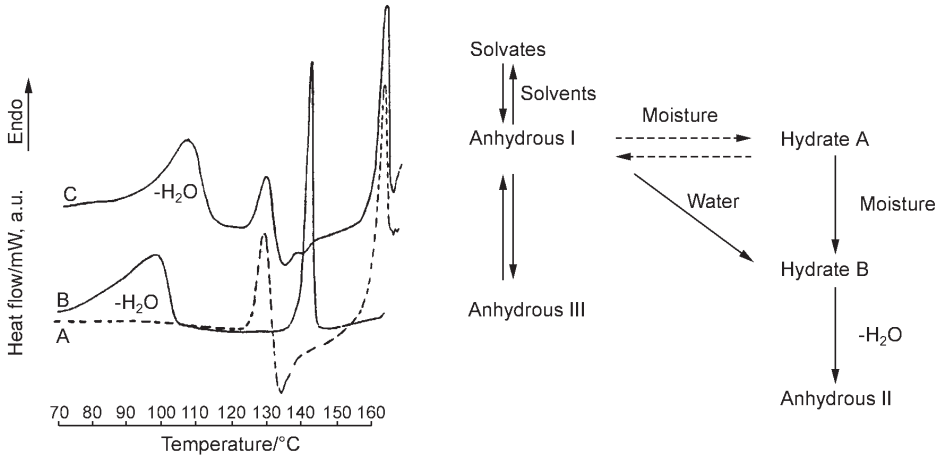


**Fig. 4** Polymorphism of hydrate DSC scans of A – initial anhydrate; B – Hydrate A; C – Hydrate B

Polymorphism of hydrates may also occur. McCauley *et al.* [17] measured the transition temperature and the crystallization conditions as well as the dehydration of two polymorphs dihydrate of a development compound. For one form the dehydration occurs directly to the anhydrate. For the other polymorph the dehydration occurs via the monohydrate. The form A of amiloride dihydrate was found more stable than form B upon milling or compression [18]. Quite recently a new polymorphic form of

the hemihydrate of aspartame as well as a 2.5 hydrate were described [19]. In the example of Fig. 4, two polymorphs of the trihydrate were identified by their different behaviour in the DSC. The DSC curve of the anhydrous form presents a dual melting.This anhydrous form is hygroscopic and transforms reversibly into an hydrate A. But if the exposition with moisture is quite longer, a second hydrate B is observed with a dehydration into a new anhydrous form. In aqueous solutions, the hydrate B, which is less soluble is obtained.

The relevance of the polymorphism of hydrates for the chemical stability is demonstrated by the example given in the Table 1 below.

**Table 1** Comparison of stability behaviour of two polymorphic forms of a monohydrate of a development new entity

| Sample | Impurities by HPLC | | |
|---|---|---|---|
| | Initial value/% | 2 weeks at 50°C/% | Exposition 1200 klux h/% |
| Monohydrate A | 1.4 | 1.4 | 11 |
| Monohydrate B | 1.0 | 12.7 | 24 |

Soustelle [20] discussed the influence of the pressure with the example of cupper sulfate. The four solid phases are the pentahydrate, the trihydrate, the monohydrate and the anhydrous form. The phase diagram $P$, $T$ has different equilibrium curves: pentahydrate to trihydrate, trihydrate to monohydrate, monohydrate to anhydrous form and also pentahydrate to the monohydrate and pentahydrate to the anhydrous form. Depending on the pressure, the TG curve of the pentahydrate can be a single dehydration process with lost of 5 molecules of water, a two steps dehydration with the intermediate monohydrate or a three steps dehydration with the intermediates trihydrate and monohydrate.

Thermal dehydration of crystalline solids hydrates in view of crystallographic structures controlling conversions has been recently reviewed by Galwey [21].

The influence of the pressure in the DSC cell is a known factor of misinterpretation of DSC curves, since depending on the pan type, the melting of the hydrate or its dehydration in the solid state may occur [3, 22]. On the other hand different results will be obtained with other techniques like thermomicroscopy or temperature resolved X-ray diffraction if the atmosphere around the sample is not the same. Han *et al.* [23] applied successfully the pressure DSC (PDSC) in order to separate the dehydration and the vaporization endotherms. The quantitation of ampicilline trihydrate in mixtures was possible. Depending on the pressure, the dehydration occurs with formation of the amorphous form or of a new polymorph of the ampicilline anhydrate. The same author proposes a humidity controlled TG and X-ray powder diffractometry for the kinetic study of the dehydration of ampicillin trihydrate, the calculation of the transition vapor pressure and the knowledge of the critical water vapor pressure above it the trihydrate is the stable solid phase [24].

*Sorption/desorption isotherms*

Studies of hydration and dehydration are generally carried out by crystallizations in water or water/organic solvent mixtures or by the measurement of water sorption and desorption isotherms. Water can be sorbed without or with phase change. Hygroscopicity and moisture adsorption kinetics of pharmaceutical solids as well as thermodynamic background have been discussed and reviewed [25, 26].

Figures 5a and 5b show the reversible sorption-desorption curves of an amorphous substance and of a very hygroscopic crystalline anhydrate turning into a monohydrate at very low relative humidity RH.

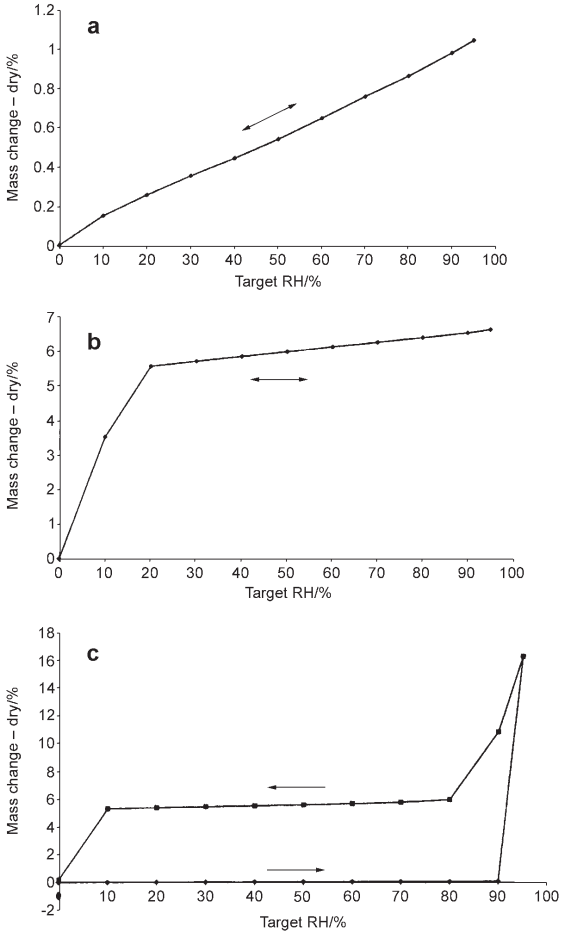


**Fig. 5** Sorption-desorption types a – reversible sorption-desorption of an amorphous sample; b – reversible sorption-desorption of a strong hygroscopic anhydrate; c – sorption-desorption of the anhydrate with formation of monohydrate and strong hysteresis of the desorption into the anhydrate

When big hysteresis are found in the desorption, as it is the case of Fig. 5c which corresponds to the study of a anhydrate, the monohydrate formed is stable enough to be analyzed separately by TG, Karl Fischer, IR, Raman, X-ray. It is then very helpful from the curves to deduce the critical relative humidity (RH) at different temperatures where the anhydrate or the hydrate have their relative stabilities. However, kinetic factors may be somewhat necessary to start the solid-solid transformation. For the example above, the transformation into the monohydrate was accelerated by seeds of the monohydrate. When the desorption is reversible, deeper analysis is necessary to conclude about the hydrate formation. Combined or coupled techniques allow to add to the calorimetric signal, informations about spectral or cristallographic data [27].

*Examples of investigations by combined methods*

The following examples will show some successful results of such investigations.

The DSC curves of the monohydrate corresponding to Fig. 5c were different if performed in a in very tight pan or with a pierced pan. In the first case, only two endotherms were observed. In the other case, the dehydration of the sample was observed in solid state, followed by two other endotherms (Fig. 6). The temperature resolved X-ray diffractions of the monohydrate show that two other anhydrates are obtained before the last transition into the stable known anhydrate (Fig. 7). The diffractograms obtained in situ allow the characterization of these metastable forms and their detection by X-ray diffraction. More complex for this drug substance was the transformation of a methanol solvate into this monohydrate after several months. X-ray diffraction was chosen for quantitation both for the drug substance and the drug product.

This example shows how hydration and dehydration may be driven by kinetic effects. Nafragel hydrochloride can crystallise as hemihydrate or as monohydrate. The sorption of the anhydrate occurs in two steps and the dehydration only in one

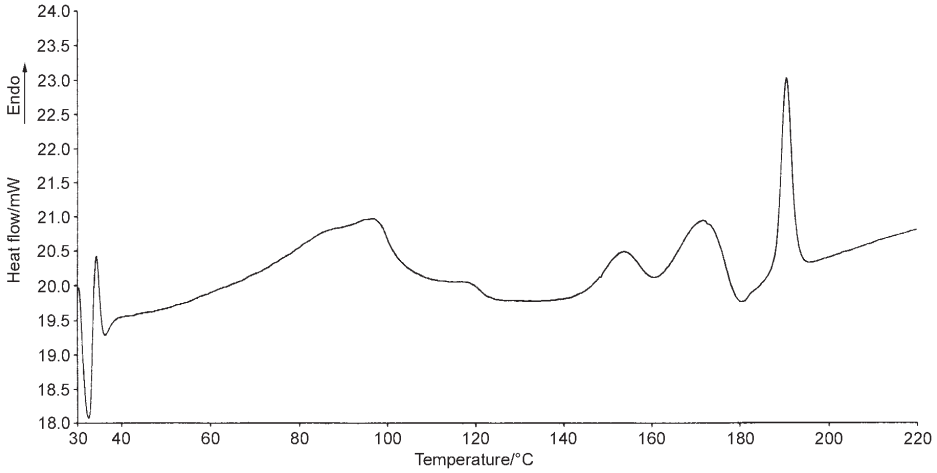


**Fig. 6** DSC curve of the monohydrate corresponding to Fig. 5c in a pierced pan

step. The analysis of hydration and dehydration at different RH, revealed different kinetics, what explains the coexistence of mixtures of both hydrated forms [28].

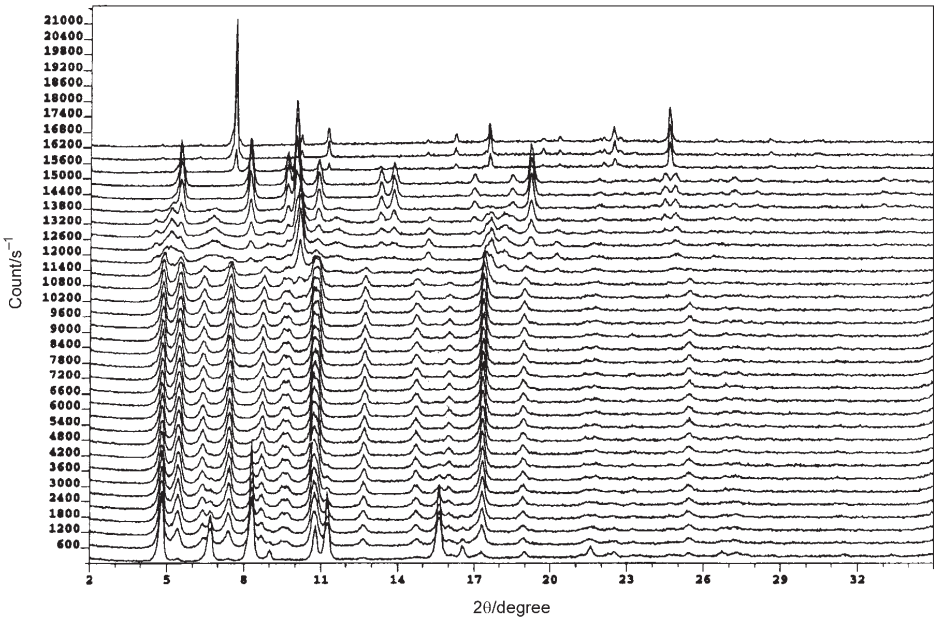


**Fig. 7** Temperature resolved X-ray diffraction of the monohydrate corresponding to Fig. 6

Figure 8a shows the reversible sorption-desorption of the first batches of a drug substance. The sub-ambient DSC analyses revealed that it was a monohydrate although already at ambient temperature and low relative humidity the crystal looses already a part of water. The studies of the X-ray diffraction at different humidity levels and at different temperatures revealed the existence of a very hygroscopic intermediate anhydrate which melts in the DSC. The single crystal structure confirmed the hydrate formation. Later on

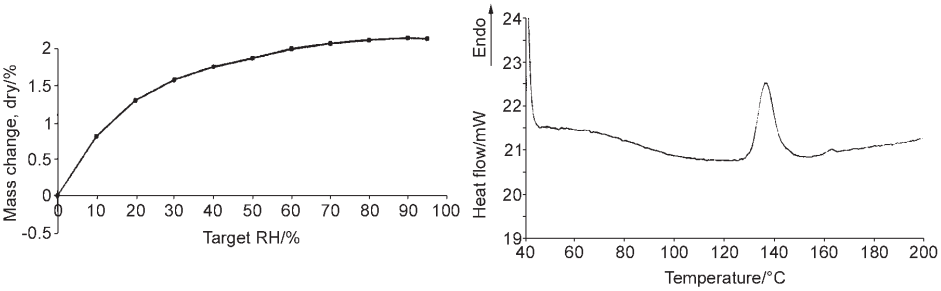


**Fig. 8** Metastable anhydrous form/Monohydrate and stable new anhydrate.
a – sorption-desorption of the metastable anhydrate/monohydrate pairs;
b – DSC curve of the monohydrate contamined with traces of anhydrous form B

in the development, a very stable anhydrous form B was found. This form is not hygroscopic and form A in presence of B transforms very quickly into the anhydrate B. Since the transformation into the anhydrate B from the melt was kinetically hindered, thermomicroscopy could be applied qualitatively. A very sensitive DSC method could be used with a LOD of approx. 0.5%. From the single crystal structures of A and B, the polymorphic purity of the reference samples of B or A could be demonstrated and the theoretical LOD of the X-ray diffraction evaluated. X-ray diffraction was the method of choice for quantitation (linearity, accuracy).

Figure 8b shows a DSC scan of the form A contaminated with traces of form B. Figure 9 shows the TG-MS of the form A. It was possible by using TG-MS to distinguish between the monohydrate and an acetone solvate of this molecule.

**Table 2** Physico-chemical properties of the anhydrous and hydrated form of a quinoline derivative. The formation of the hydrated form (form A) is driven by the water activity of the solvents

| | Method/ Conditions | Results form A, batch 1 | Results form B, batch 2 |
|---|---|---|---|
| Purity (eutectic) | DSC/1 K $min^{-1}$ | 99.8 mol% | 99.8 mol% |
| DSC melting onset | DSC/10 K$min^{-1}$ | 208°C | 195°C |
| DSC melting enthalpy | DSC/10 K $min^{-1}$ | 79.1 J $g^{-1}$ | 82.8 J $g^{-1}$ |
| Heat of solution (in MeOH) | microcalorimetry | 23.2±0.1 J $g^{-1}$ | 25.1±0.1 J $g^{-1}$ |
| Thermogravimetry | | 0.5% | <0.05% |
| Crystallinity | XRPD | high | high |
| Dissolution rate in: a) 0.1 N HCl+0.5%Tween 20 | Flow cell $T$=25°C | | |
| $t_{50\%}$ | | 5 min 22 s/ 6 min 11 s | 4 min 4 s/ 5 min 17 s |
| $t_{80\%}$ | | 10 min 57 s/ 12 min 53 s | 10 min 36 s/ 11 min 50 s |
| $t_{90\%}$ | | 16 min 20 s/ 17 min 56 s | 18 min 40 s/ 19 min 10 s |
| b) water+0.2% LDAO | | | |
| $t_{50\%}$ | | 7 min 09 s/ 10 min 23 s | 11 min 47 s/ 9 min 19 s |
| $t_{80\%}$ | | 22 min 56 s/ 24 min 44 s | 28 min 35 s/ 27 min 56 s |
| $t_{90\%}$ | | 42 min 09 s/ 38 min 26 s | 48 min 53 s/ 50 min 21 s |
| Second DSC run after melting | DSC | $T_g$=92.5°C | $T_g$=93.9°C |

The thermodynamic stability of hydrated forms in mixed solvents depends on water activities [29–31] as demonstrated in the figure 10a for a quinoline derivative. The modification B is anhydrous. Modification A is an hydrated form as demonstrated by the study

of the sub-ambient DSC. The corresponding sorption-desorption isotherm (Fig. 10b) is very similar to the example of Fig. 8. The formation of A or B is driven by the water activity of the solvents. Form B was selected. The detection of form A was monitored by X-ray diffraction and TG. The Table 2 below summarizes some results.

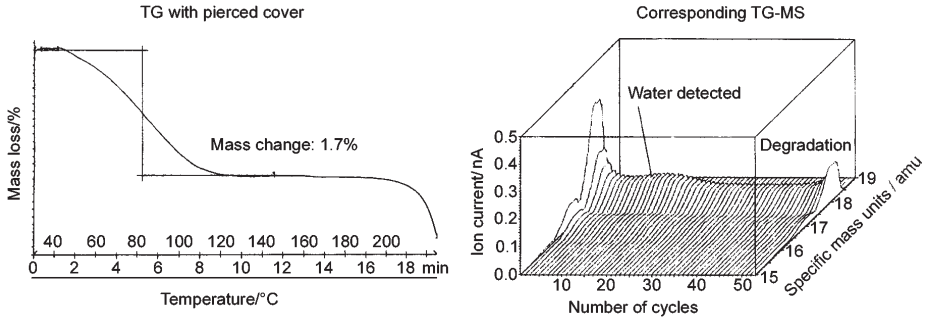


**Fig. 9** TG-MS of the monohydrate of Fig. 8

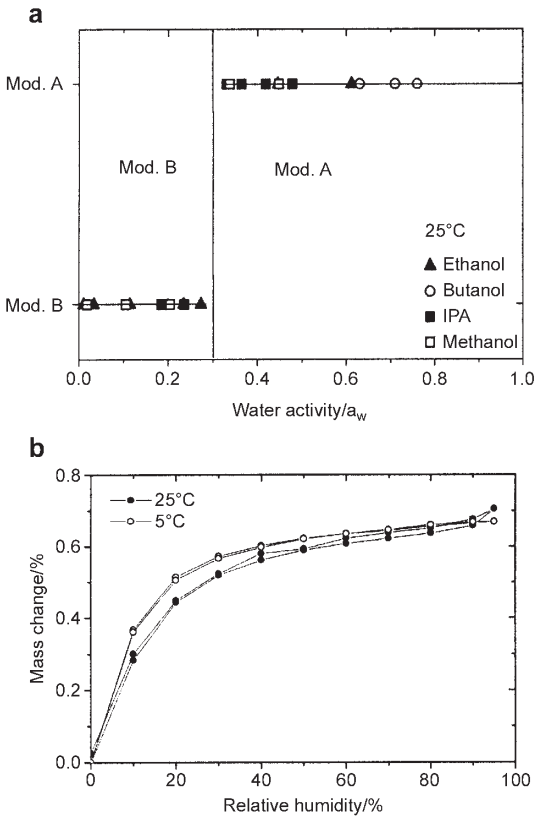


**Fig. 10** Influence of water activity in solvents of crystallization a – diagram *vs.* water activity; b – sorption of form A

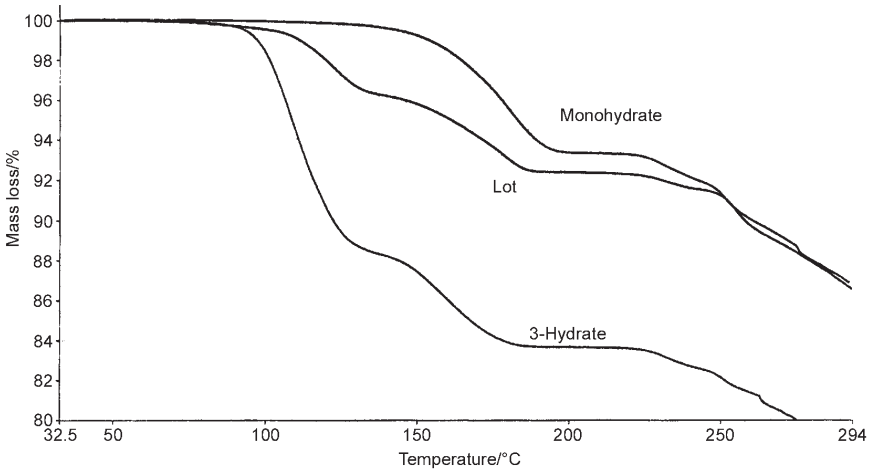


**Fig. 11** TG curves of a monohydrate, of a trihydrate and of a lot containing monohydrate and trihydrate

The next example is a drug substance developed as a monohydrate. This hydrated form is manufactured in ethanol/water. The thermogravimetric analysis of a lot obtained in a scale-up revealed a significant difference (Fig. 11), although results of Karl Fischer were within requirements. Crystallization experiments, equilibration in solvent mixtures, TG, DSC, X-ray diffraction, sorption and desorption as well as combined techniques were used for a deeper investigation. Besides the anhydrate and the monohydrate, a trihydrate is obtained. The solubilities in water are summarized in Table 3.

**Table 3** Solubility behaviour of a drug substance in water at different temperatures and areas corresponding to anhydrate, monohydrate and trihydrate

| Temperature/°C | Solubility/mg $mL^{-1}$ | Residual solid |
|---|---|---|
| 10 | 2.1 | trihydrate |
| 25 | 2.8 | trihydrate |
| 40 | 10.9 | trihydrate |
| 60 | 25.3 | monohydrate |
| 80 | 31.6 | anhydrate |

The dehydration of the trihydrate occurs via the monohydrate (Fig. 11). The monohydrate is not hygroscopic. Through drying at 80°C, the anhydrate obtained is hygroscopic and transforms into the monohydrate (Fig. 12).

Manufacturing conditions of the monohydrate were optimized and a quantitative X-ray diffraction method was developped.

For the last example, laboratory batches were strongly affected by grey impurities. By using an aqueous mixture, the impurities were removed and after drying a

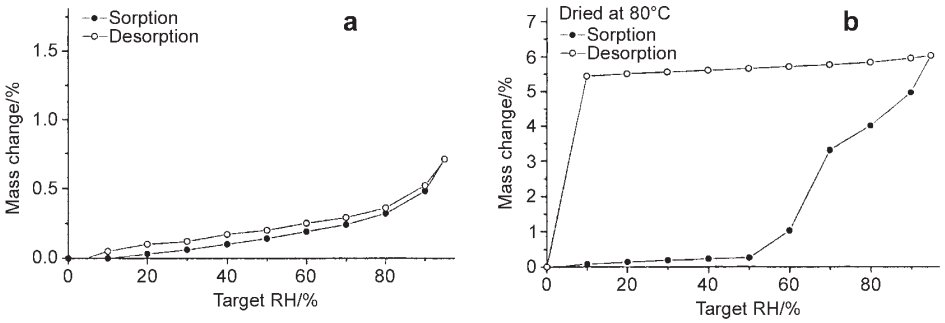


**Fig. 12** Sorption-desorption of the monohydrate (left) of Fig. 11 and of the corresponding anhydrate (right) obtained by drying at 80°C

new polymorph was obtained. This form was highly hygroscopic and converted into a trihydrate. Since the drug substance was chemically very sensitive to moisture, it was decided to manufacture the anhydrous form. Slurry of the purified trihydrate in methanol allowed the transformation into the stable white anhydrous form.

## Conclusions

Hydrate formation leads to complex behaviour of drug substance and drug products in every step of processing and after storage. Adequate investigations are necessary in very early stage of development for the proper choice of the candidate form, the choice of the formulation, of the process and of the packaging. A deep insight needs several analytical complementary techniques with a high level of information. Particurlarly informative are X-ray diffraction experiments in chambers of different humidity in parallel to sorption-desorption isotherms. Dehydration studies by TG under different temperatures and pressure simulate drying and milling processes. The crystal stucture obtained by single crystal diffraction or by computational calculation from X-ray diffractometry allows to understand the type of the bonds between water and drug substance. Modelling capabilities are extremely helpful as an help for quantitative methods in drug substance and drug products.

## References

1 J. K. Haleblian and W. J. McCrone, Pharm. Sci., 58 (1969) 911.
2 J. K. Haleblian, Pharm. Sci., 64 (1975) 1269.
3 D. Giron, Thermochim. Acta, 248 (1995) 1.
4 D. Giron, Labo-Pharma-Probl. Techn., 307 (1981) 151.
5 D. Giron, S.T.P. Pharma, 4 (1988) 330.
6 J. Bernstein, R. J. Davey and J. O. Henck, Angew. Chem. Int. Ed., 38 (1999) 3340.
7 K. R. Morris, Structural aspects of hydrates and solvates. (Polymorphism in Pharmaceutical Solids Brittain H. G. ed., Marcel Dekker, New York), Drugs Pharm. Sci., 95 (1999)125.

8 R. K. Kankhari and D. J. W. Grant, Thermochim. Acta, 248 (1995) 61.
9 E. Suzuki, Chem. Pharm. Bull., 37 (1989) 493.
10 H. Ando, Drug Dev. Ind. Pharm., 21 (1995) 2227.
11 C. M. Adeyeye, Int. J. Pharm., 116 (1995) 65.
12 D. Giron, S.T.P. Pharma (Hors serie), 6 (1990) 87.
13 J. T. Carstensen, Drug Dev. Ind. Pharm., 14 (1988) 1927.
14 International Conference on Harmonization (ICH) Guideline Specification Q6A, Decision Tree: Investigating the need to set acceptance criteria for polymorphism in drug substances and drug products, 1999.
15 P. Di Martino, F. Piva, P. Conflant and A. M. Guyot-Hermann, J. Therm. Anal. Cal., 57 (1999) 95.
16 D. Giron and C. Goldbronn, J. Thermal Anal., 49 (1997) 907 and J. Therm. Anal. Cal., 51 (1998) 727.
17 J. A. McCauley, R. J. Varsolona and D. A. Levorse, J. Phys. D: Appl. Phys., 26 (1993) B85.
18 M. J. Jozwiakowski, S. O. Williams and R. D. Hathaway, Int. J. Pharm., 91 (1993) 195.
19 S. S. Leung, B. E. Padden, E. J. Munson and D. J. W. Grant, J. Pharm. Sci., 87 (1998) 501.
20 M. Soustelle, Handbook of Powder Technology, J. C. Williams and T. Allen, Eds, Vol. 9, Powder Technology and Pharmaceutical Processes, D. Chulia, M. Deleuil and Y. Pourcelot, Eds, Elsevier 1994, p. 27.
21 A. K. Galwey, Thermochim. Acta, 355 (2000) 181.
22 D. Giron, Acta Pharm. Jugosl., 40 (1990) 95.
23 J. Han, S. Gupte and R. Suryanarayanan, Int. J. Pharm., 170 (1998) 63.
24 J. Han and R. Suryanarayanan, Thermochim. Acta, 329 (1999) 163.
25 K. Umprayn and R. W. Mendes, Drug Dev. Ind. Pharm., 13 (1987) 653.
26 M. J. Kontny and G. Zografi, Drugs Pharm. Sci., 70 (1995) 387.
27 D. Giron, J. Therm. Anal. Cal., 64 (2001) 37.
28 H. Kitaoka, Chem. Pharm. Bull., 43 (1995) 1744.
29 P. L. Gould, J. R. Howard and G. A. Oldershaw, Int. J. Pharm., 51 (1989) 195.
30 H. Zhu, C. Yuen and D. J. W. Grant, Int. J. Pharm., 135 (1996) 151.
31 H. Zhu and D. J. W. Grant, Int. J. Pharm., 139 (1996) 33.