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Thermochimica Acta 340–341 (1999) 117–129

thermochimica acta

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Physico-chemical characterisation of different solid forms of spironolactone

V. Berbenni[a,*], A. Marini[a], G. Bruni[a], A. Maggioni[a],
R. Riccardi[a], A. Orlandi[b]

[a]*C.S.G.I, Dipartimento di Chimica Fisica dell'Università di Pavia-Viale, Taramelli 16-27100, Pavia, Italy*
[b]*Glaxo Wellcome, Medicines Research Centre, Via Fleming 2-37135, Verona, Italy*

Accepted 6 August 1999

Abstract

Spironolactone is a steroidal diuretic showing incomplete oral behaviour because of its low solubility and slow dissolution rate. Four solid forms were obtained by recrystallisation of a commercial sample from different solvents (acetone, methanol, ethyl acetate). These samples have been characterised by Fourier transform infrared (FT-IR) Spectroscopy, X-ray powder diffraction, thermogravimetric analysis coupled with FT-IR spectroscopy (TG/FT-IR) and differential scanning calorimetry (DSC). The aim of the work is establish evidence for differences, if any, between the forms and, possibly, to understand their nature. A careful analysis of the collected data makes it possible to rule out the presence of any solvate and to distinguish three different polymorphs. © 1999 Elsevier Science B.V. All rights reserved.

Keywords: Spironolactone; Ethyl acetate; Acetone

1. Introduction

Spironolactone (see Fig. 1) is a steroidal diuretic showing variable and incomplete oral behaviour because of its low water solubility and slow dissolution rate [1]. However it is well known that different crystal phases of the same molecule may have different solubility, dissolution rate and, as a consequence, different bioavailability. As is reported in literature, different solid forms of spironolactone have been prepared by recrystallisation from various solvents [2], such as acetonitrile, ethyl acetate or ethanol. According to some authors [3] the ability of spironolactone to recrystallise in different forms is due to its molecular flexibility. As a matter of fact, a thorough structural study performed on single crystal specimens obtained by different solvents [4] established evidence for two polymorphic forms and four solvates. On the other hand, Neville et al. [5–7] distinguished some spironolactone solvates by Fourier transform infrared (FT-IR) diffuse reflectance spectroscopy and characterised them by thermal analysis (TGA, DSC). However, there is poor agreement in the literature on important aspects such as the number and nature of the solvates [2–4] and the ability of IR spectroscopy to discriminate between different polymorphs [5]. In the present work some forms of spironolactone, namely those prepared by methanol, acetone and ethyl acetate (two different preparation routes — see Section 2 for details) have been thoroughly characterised compar-

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[*] Corresponding author.

PII: S0040-6031(99)00258-0

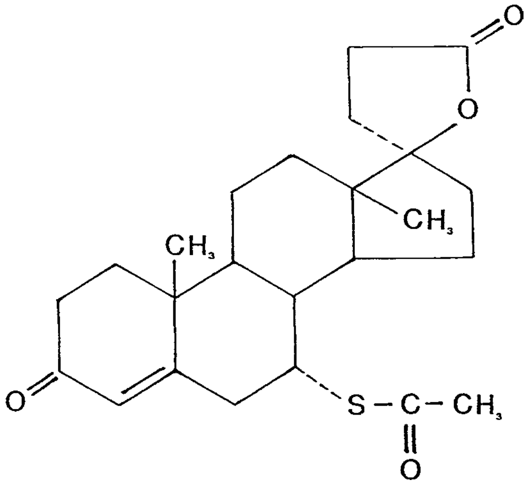

Fig. 1. Spironolactone molecule.

ing the results obtained by FT-IR, XRD, TG/FT-IR and DSC in order to put into evidence the differences between the samples.

2. Experimental

2.1. Products

All samples have been prepared starting from a commercial product (Aldrich Chimica, Italy, purity 99.97%). The precursor powder has been dissolved in methanol (Fluka Chimica puriss., Italy), acetone (Fluka Chimica puriss., Italy) and ethyl acetate (Fluka Chimica puriss., Italy) to obtain saturated solutions at nearly the boiling point of each solvent. Then, after filtration, the solutions were slowly cooled down to room temperature (RT). When working with ethyl acetate a different route was also followed: a saturated solution was prepared at RT, after filtration the solution was left at RT for several days to undergo spontaneous evaporation until a solid crystallised. In the following, the different forms obtained will be referred to as M (methanol), A (acetone), E (ethyl acetate) and EV (ethyl acetate, RT evaporation).

2.2. Apparatus and procedures

FT-IR measurements were recorded by an FT-IR spectrometer (Nicolet 730) fitted by a diffuse reflectance attachment (Drift Collector by Spectratech). Spectra (4000–400 cm^{-1}) were obtained by FFT of 64 coadded interferograms (resolution 2 cm^{-1}) collected at RT and with a dry nitrogen flow of 15 l/min purging the spectrometer. The samples were prepared as physical mixtures of about 5% w/w spironolactone in potassium bromide. Pure potassium bromide was used as background.

XRD patterns were collected with a Philips PW1710 powder diffractometer fitted with a vertical goniometer (Philips PW 1050). CuKα radiation monochromatised by a bent graphite crystal was used. The measurements were made on small samples (about 20 mg) sandwiched between two polyethylene films so as to get sample thickness of about 0.1 mm.

TGA measurements were carried out in a TGA Analyser (DuPont 951) connected to a TA 2000 System (TA Instruments). The samples (about 3–4 mg) were placed in a Pt holder and heated up to the final temperature ($\approx$370°C) under a dry nitrogen flux of 80 ml/min and at a heating rate of 5°C/min. TG/FT-IR measurements were carried out by connecting a gas cell, placed inside the FT-IR spectrometer and maintained at 290°C, to the TGA Analyser. At the end of the measurements the spectral data (spectral range 4000–500 cm^{-1}, 16 coadded interferograms, resolution 16 cm^{-1}, DTGS detector) were treated by SID Nicolet Proprietary Software to yield the Gram–Schmidt reconstruction and generate spectra corresponding to the different TGA temperatures.[1]

DSC measurements were performed with a heat flux DSC cell (DuPont 910) also connected to the TA 2000 Thermal Analysis System. The samples (about 3–4 mg) were put in aluminium pans fitted with pierced lids. The cell was flushed with dry nitrogen (50 ml/min). Different heating rates (0.5–1–2–5 and 10°C/min) were used. The samples were heated up to a temperature just above the melting peak.

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[1] The Gram–Schmidt reconstructions (GSR curves in the following) reflect the shift of a definite number of points of the centerburst of the interferograms due to the gases released in the TGA and entering the IR cell. It represents the total IR signal and does not correspond to any specific IR absorber. In principle the GSR curve should be superimposed on the DTG curve [8].

3. Results and discussion

3.1. DRIFT measurements

Fig. 2 reports the IR spectrum of a sample recrystallised from methanol. The same pattern, in the following referred to as type 1 spectrum, is displayed by both ethyl acetate derived samples. Fig. 3 shows the spectrum of a sample recrystallised from acetone that in the following will be referred to as type 2 spectrum.

The fact that IR Diffuse Reflectance distinguishes only two forms of spironolactone is clearly repre-

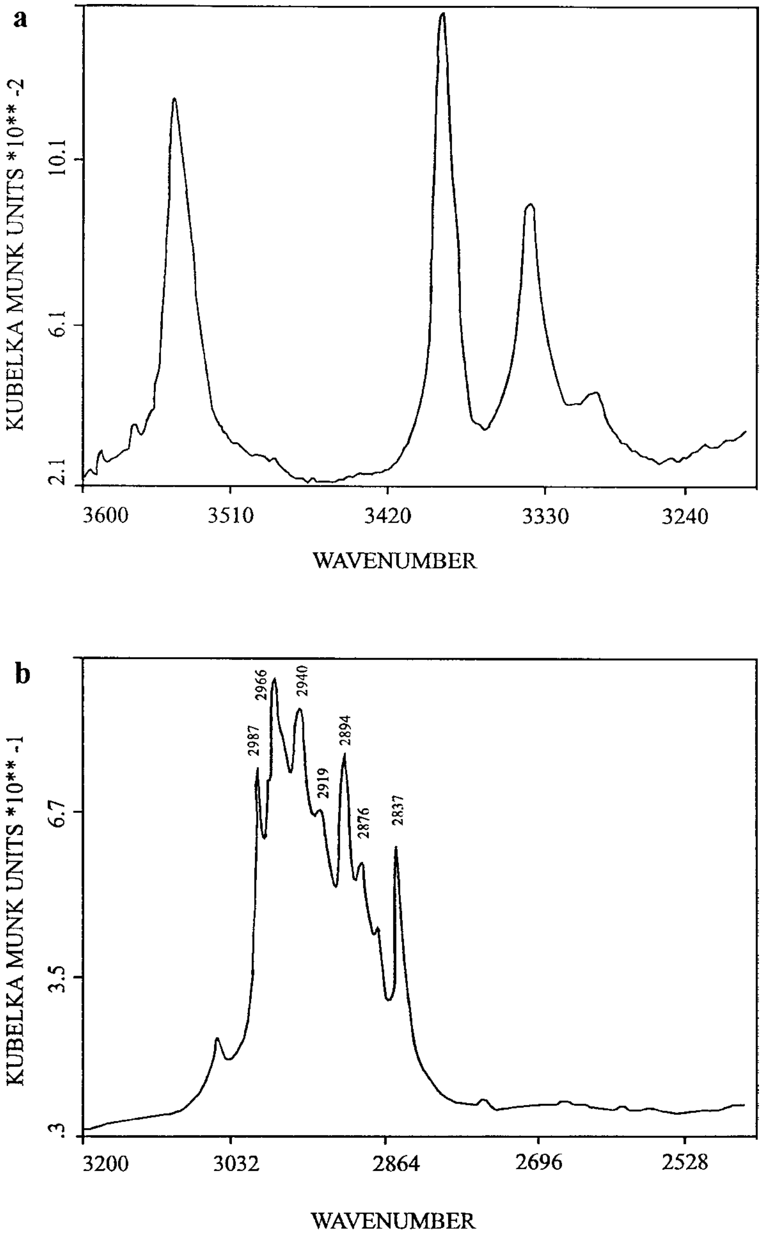


Fig. 2. Type "1" DRIFT spectrum (M, E, EV samples). Parts a, b, c and d report different spectral regions.

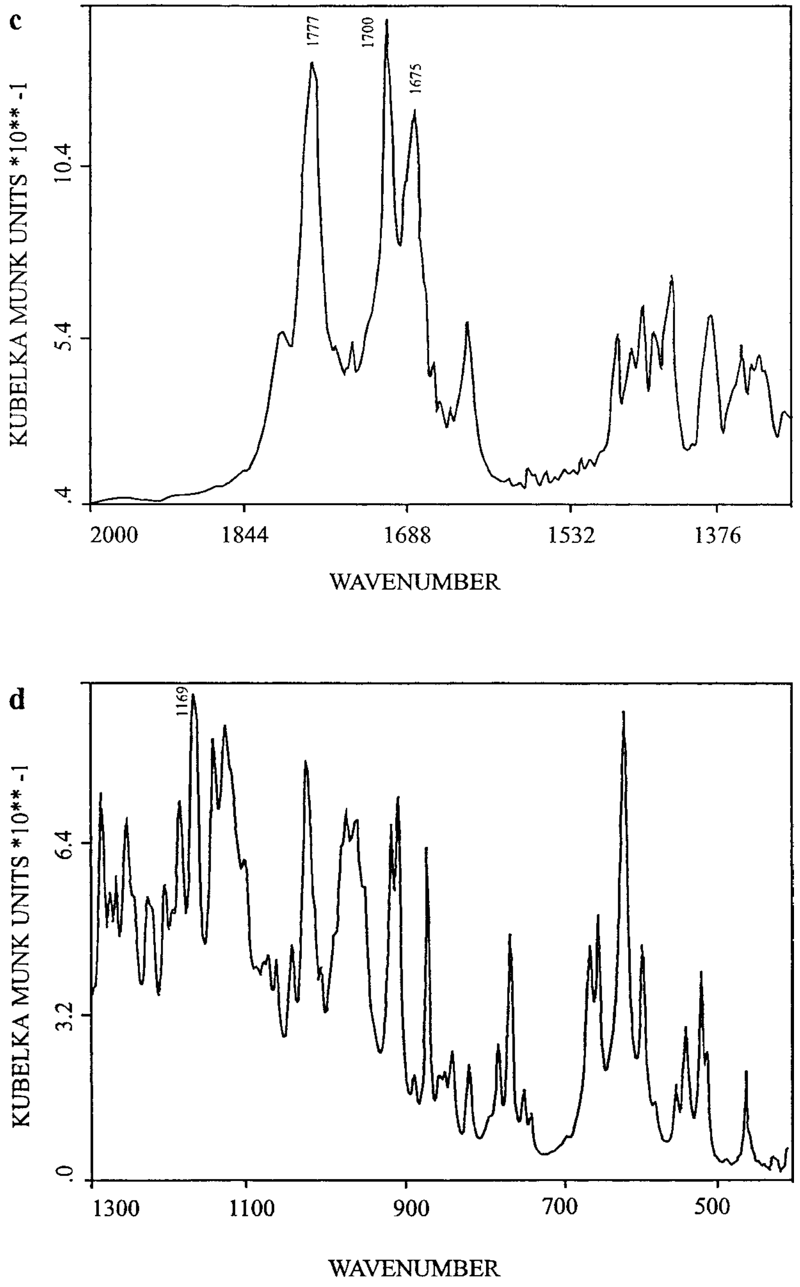


Fig. 2. (*Continued*)

sented in Table 1, that reports the wavenumbers of overtones and combination bands [5] in the 3600–3200 cm^{-1} region. It can be seen that only two types of vibrational behaviour can be distinguished in the four samples examined.

Our spectra of M, E and EV samples do show neither the ≈3500 cm^{-1} broad band (related to the stretching vibration of hydroxyl group) nor the ≈1720 cm^{-1} band (related to the presence of ester carbonyl) which, on the contrary, were present in the spectra of Neville et al. [6]. Our findings suggest that neither methanol nor ethyl acetate solvents are retained in the pertinent recrystallised samples. The reason for the disagreement could be that the samples of Neville et al. were recrystallised in a different temperature range.

Coming to the differences between the two types of spectrum it can be said that:

- the stretching region of the alkane residues is more resolved in type 1 spectrum only (see Fig. 2b and Fig. 3b);

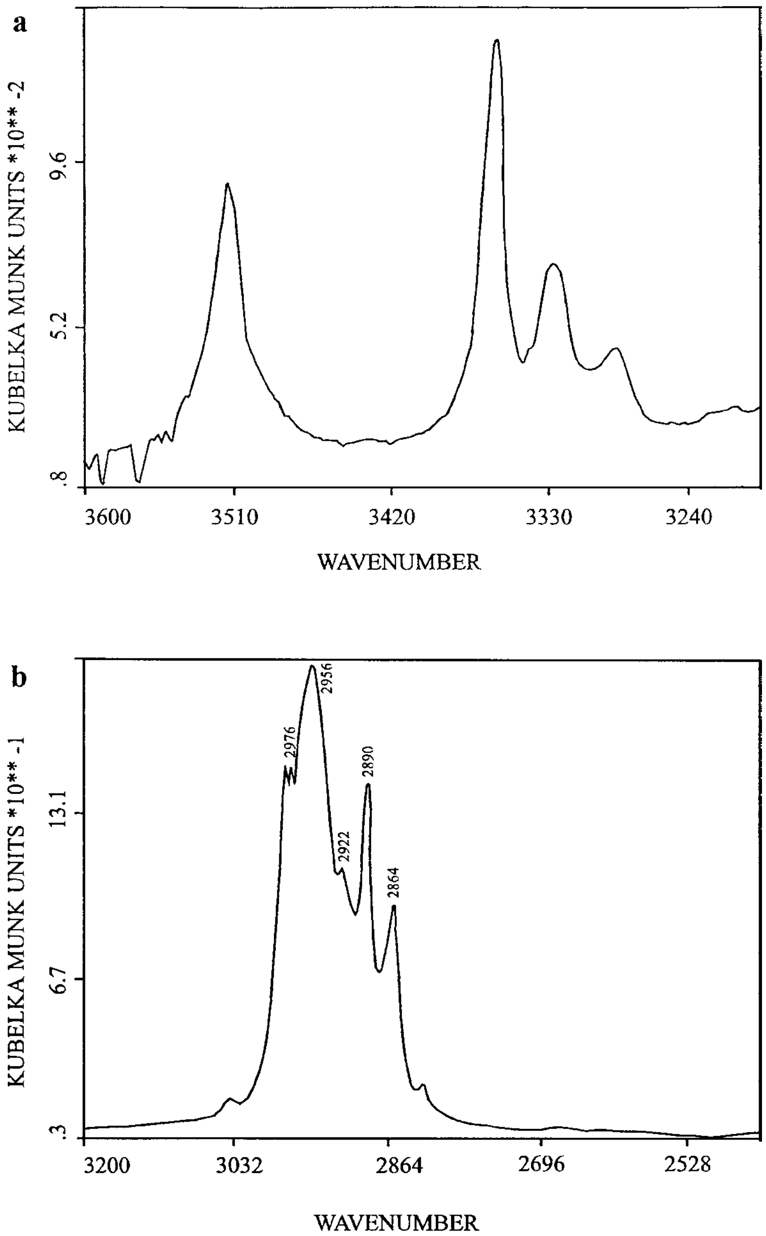
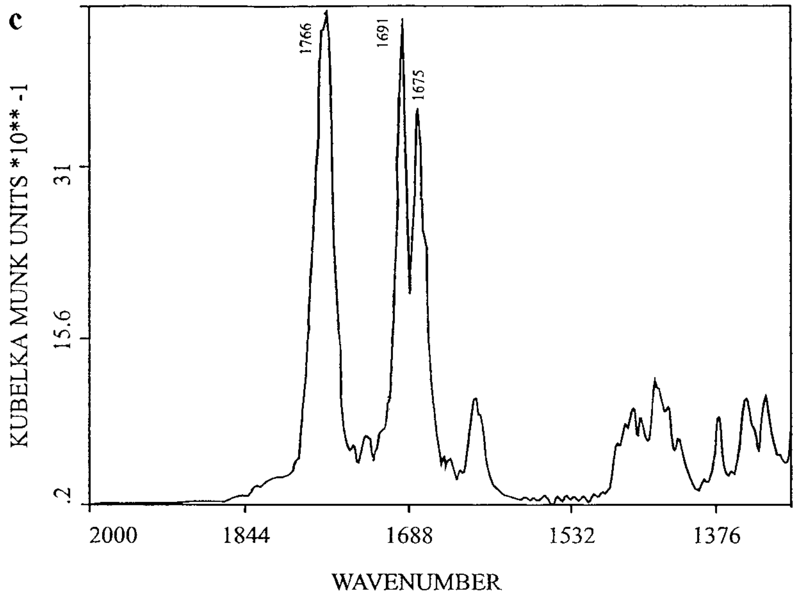


Fig. 3. Type "2" DRIFT spectrum (A sample). Parts a, b, c and d report different spectral regions.



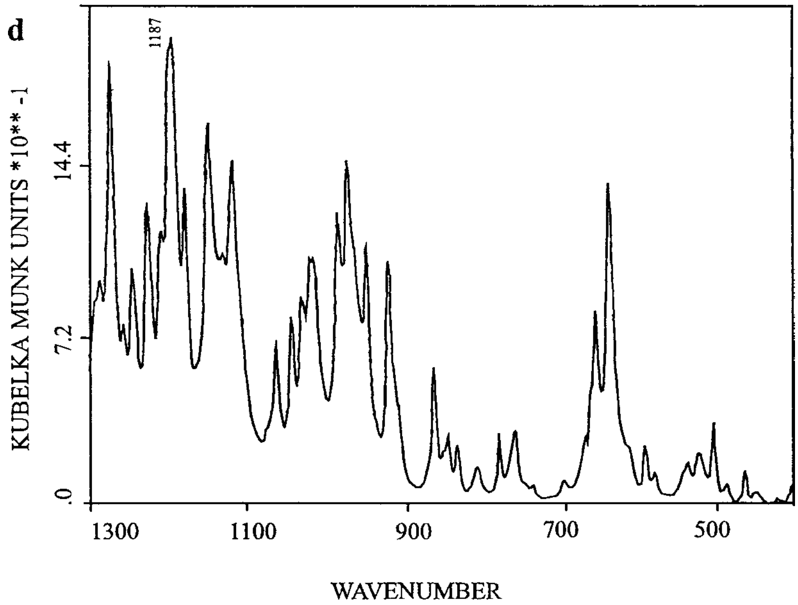


Fig. 3. (*Continued*)

- both type 1 and 2 spectra show the peak of the α–β unsaturated cyclic ketone at 1675 $\mathrm{cm^{-1}}$ (Fig. 2c and Fig. 3c);
- the stretching vibration of the thioacetylic function (reference 1700–1680 $\mathrm{cm^{-1}}$) is at 1691 $\mathrm{cm^{-1}}$ in spectrum 2 (Fig. 3c) while it is shifted to 1700 $\mathrm{cm^{-1}}$ in spectrum 1 (Fig. 2c);
- the γ-lactone carbonyl peak is at 1777 $\mathrm{cm^{-1}}$ in spectrum 1 while it is located at 1766 $\mathrm{cm^{-1}}$ in spectrum 2. Furthermore spectrum 1 shows

Table 1
Overtones and combination bands[a]

Sample	Spectrum type	1	2	3	4
M	1	3536	3381	3330	3285 (vw)
E	1	3536	3381 (sh: 3361)	3329	3285 (vw)
EV	1	3536	3381	3331	3285 (vw)
A	2	3513	3361	3323	3285

[a] Wavenumbers: cm^{-1}; vw = very weak; sh = shoulder.

a shoulder towards higher frequencies (1805 cm^{-1});

- the patterns of the stretching vibration of the ester C–O group (reference 1370–1160 cm^{-1}) are different for the two types of spectrum. In type 2 spectrum, a candidate peak is at 1187 cm^{-1} (the stronger peak in the fingerprint region, Fig. 3d). In type 1 spectrum, a peak is present at 1186 cm^{-1} but is of lower intensity, the stronger peak being now at 1169 cm^{-1} (Fig. 2d). The 1266 cm^{-1} peak, which is present in spectrum 2, is greatly diminished, a doublet being present instead at 1267–1255 cm^{-1};
- the C–S bond vibration (reference 670 ± 35 cm^{-1}, weak to medium intensity) is present as a double peak at 656 and 639 cm^{-1} (the stronger component) in spectrum 2 (Fig. 3d). In spectrum 1 the 656 cm^{-1} peak is split into a doublet (662 and 652 cm^{-1}) while the 638 cm^{-1} one is substituted for by a much stronger peak located at 621 cm^{-1}. It can be said, as a conclusion, that both γ-lactone cyclopentane ring and thioacetylic function are in a more strained conformation in M, E and EV samples (spectrum 1). The difference in the conformation of the thioacetylic group is confirmed by the different vibrational features of the C–S group in the two types of spectrum. Indeed such a vibration is known to be sensibly affected by the molecular conformation [5].

3.2. XRD measurements

Fig. 4 reports the XRD spectra of all samples. The broad and weak effects of the EV pattern (Fig. 4b) suggest that this sample contained particles of quite a small size. However, the patterns of A, E and M samples show differences both in the angular position of some peaks and in the relative intensities of their common reflections. Thus it can be stated that the XRD measurements recognise at least three different polymorphic forms of spironolactone. Note, however, that some differences in the relative intensities could be due to preferred orientation effects (that are common in organic substances), rather than to real structural differences.

3.3. TG/FT-IR measurements

Fig. 5a–d reports the Gram–Schmidt spectral reconstruction curves (GSR in the following) along with the corresponding DTG curves(derivative thermogravimetry). It appears at first sight that the A sample (Fig. 5b) is the only one where a single peak is present in the relevant GSR curve. This confirms that the A sample is different from the other ones (see FT-IR and XRD results).

Table 2 reports the onset (T_0) and the final ($T_{fin,I}$) temperatures of the first GSR peak along with the corresponding mass changes over the peak (ΔM_I). Also reported are the mass loss values at the end of the run (ΔM_{fin}).

No sensible differences seem to exist between the E, EV and A forms with respect to both mass losses (ΔM_I and ΔM_{fin}). On the contrary the M sample shows a higher value of both ΔM_I and ΔM_{fin} and the highest

Table 2
TG/FT-IR data[a]

Sample	T_0 (°C)	$T_{fin,I}$ (°C)	ΔM_I (%)	ΔM_{fin} (%)
M	240	305	50.0	94.0
E	206	312	23.7	80.0
EV	206	315	25.5	76.0
A	233	310	26.0	75.0

[a] T_0 and $T_{fin,I}$ are the initial and final temperatures of the first GSR peak, ΔM_I and ΔM_{fin} the mass losses corresponding to the first peak and to the final point.

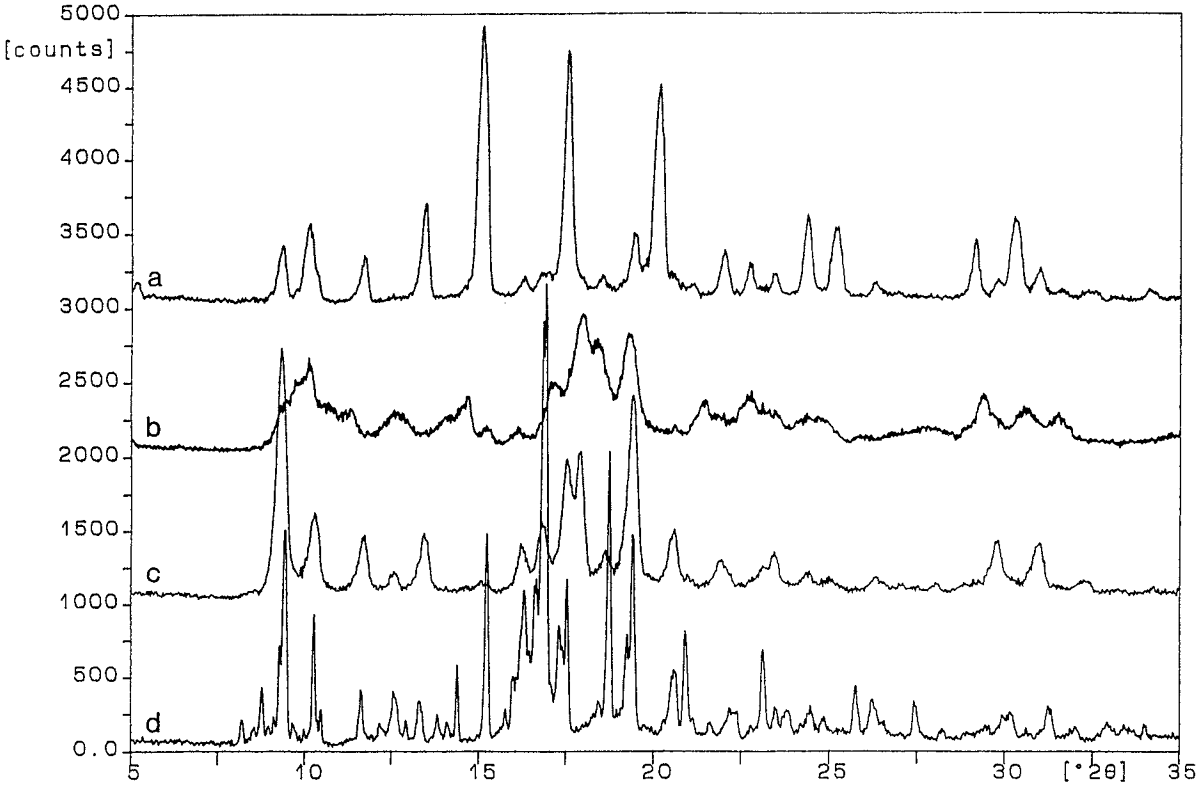


Fig. 4. XRD spectra of the solid forms: (a) M, (b) EV, (c) E, (d) A.

decomposition temperature ($T_0 = 240°C$). Thus it seems that the M sample can be distinguished from the other ones by a different thermal stability. Indeed, by examining the IR spectra of the gas evolved under the first GSR peak, it can be easily seen that the M form evaporates as it is (Fig. 6) while the other forms release molecular fragments containing sulphur (CH_3COSH or COS) (Fig. 7) besides water and carbon dioxide (perhaps due to an oxidative degradation process). As the A sample has been distinguished from all the other ones both by FT-IR and XRD, it can be concluded that TG/FT-IR measurements enable sample M to be distinguished from all other forms on the basis of its higher thermal stability and its different evaporation/decomposition pattern.

3.4. DSC measurements

Table 3 reports the melting enthalpies of the samples, along with the onset and maximum temperatures,from DSC experiments carried out at different heating rates. It should be noted that in no case spironolactone melting has been detected in a second heating run.

It can be seen that for DSC too, the A sample is different from all the others (it shows the lowest values of melting enthalpies and peak temperatures). A common feature of the A and EV samples is that their melting enthalpies decreases with decrease in the heating rate. This trend can be explained by admitting that these samples undergo a partial decomposition before fusion. The extent of such a decomposition is different for the two forms and clearly increases, though with different rates, by decreasing heating rate (a low heating rate allows more time for the occurrence of such solid state decomposition). Work is in progress [9] to fully understand the nature of the decomposition process and to account for its effect on the melting enthalpy. On the contrary, no appreciable dependence of the melting enthalpy on the heat-

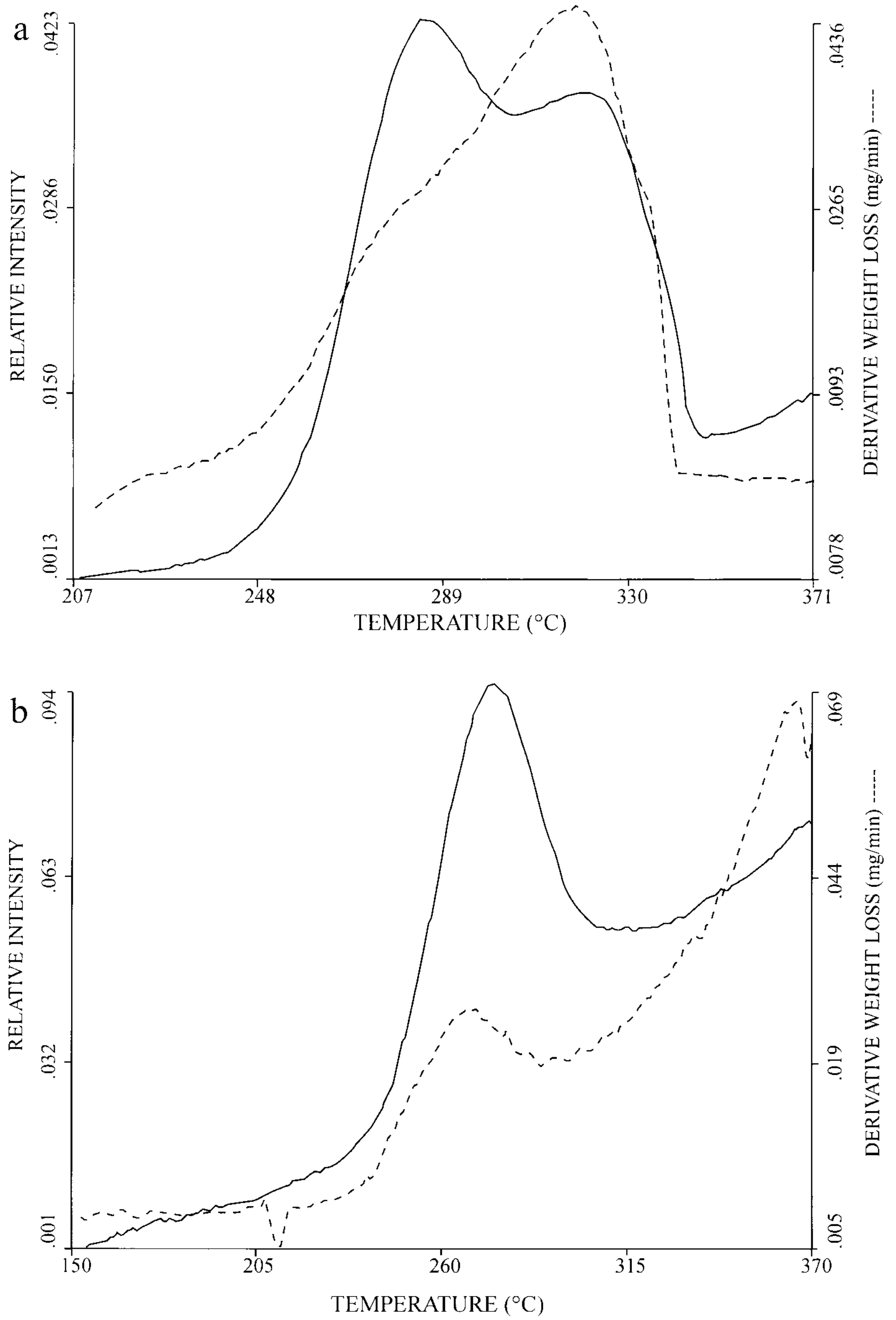


Fig. 5. GSR (solid lines) and DTG (broken lines) curves of the different solid forms (heating rate: 5°C/min). (a) M, (b) A, (c): E, (d): EV.

ing rate is shown by the M and E samples. As their melting enthalpies (mean values over all the heating rates: 52.6 ± 1.5 J/g for M sample and 50.6 ± 3.0 J/g for E sample) are the same within experimental error and their peak temperatures are quite similar, the conclusion can be drawn that the M and E samples

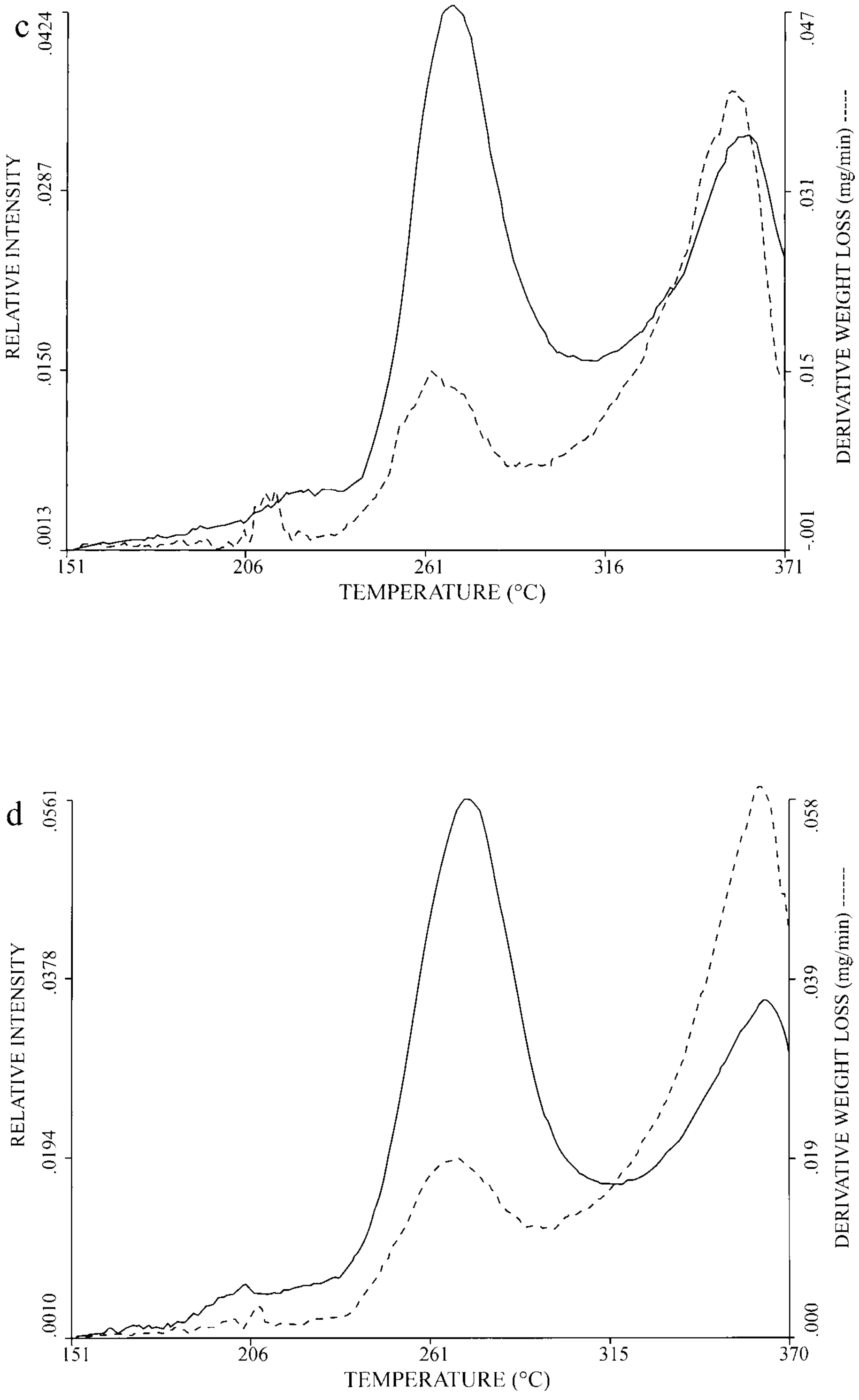


Fig. 5. (*Continued*)

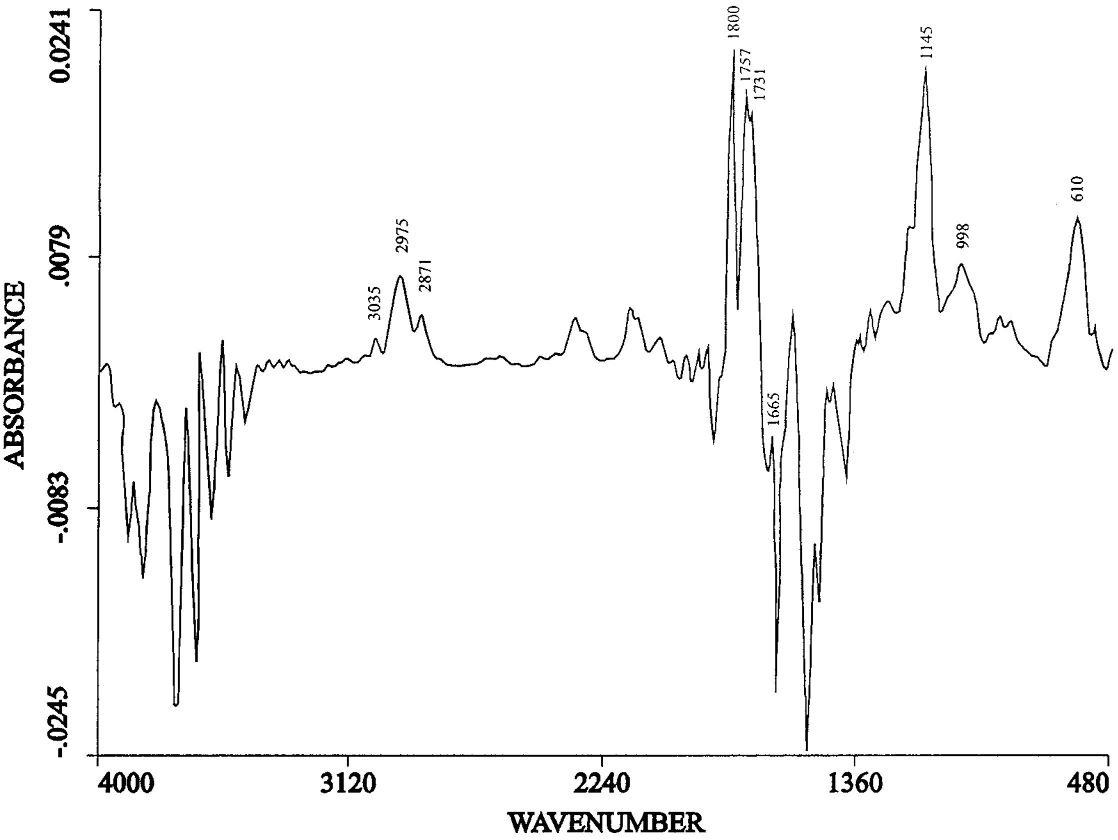


Fig. 6. FT-IR spectrum of the gas evolved by M sample near the maximum of the first GSR peak (from TG/FT-IR).

Table 3
DSC results on the different samples[a]

β	T_0 (°C)				T_{max} (°C)				$\Delta_{fus}H$ (J/g)			
	A	M	E	EV	A	M	E	EV	A	M	E	EV
10.0	197.5	211.4	210.0	207.2	205.2	212.8	211.7	209.9	39.5	53.4	50.9	49.9
5.0	195.2	209.9	208.8	203.5	201.9	212.3	210.3	206.7	33.2	53.7	46.5	47.7
2.0	190.3	209.0	206.8	200.1	197.6	210.5	209.1	204.4	27.9	51.4	54.3	44.0
1.0	183.9	208.7	205.2	194.9	191.5	210.1	208.1	199.8	24.8	53.8	49.1	36.9
0.5	179.1	205.4	203.7	192.9	187.9	207.2	206.8	197.8	22.2	50.5	52.3	29.4

[a] β is the heating rate (°C/min), T_0, T_{max} the extrapolated onset and maximum temperatures, and $\Delta_{fus}H$ the melting enthalpy. Note that all the measurements have been run in duplicate.

cannot be distinguished by DSC. Then, the new information brought about by DSC measurements is that the E and EV samples have different thermal stability before melting.

4. Conclusions

According to the collected experimental evidence the following conclusions can be made:

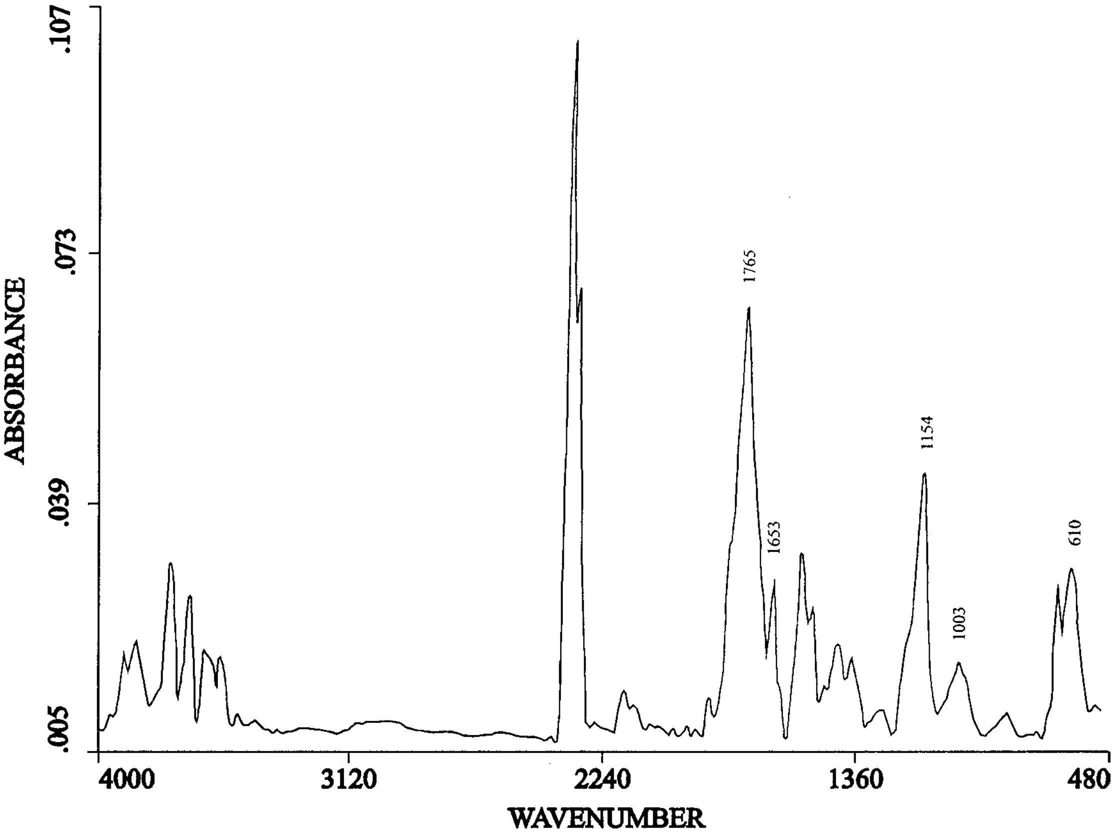


Fig. 7. FT-IR spectrum of the gas evolved by A sample near the maximum of the first GSR peak (from TG/FT-IR).

- the A sample is the only form that is shown to be different from the other ones by all the experimental techniques. Actually it is the only form that shows a type "2" infrared spectrum (see FT-IR evidence), besides having its own diffraction pattern (see XRD evidence) and its different melting enthalpy (see DSC evidence). No further observations are needed to distinguish the A sample, which constitutes a polymorphic form of spironolactone (form A);
- the DSC evidence shows a difference between the two ethyl acetate samples and suggests that the EV one undergoes partial decomposition before melting. This fact does not reflect a difference between the E and EV samples at a molecular level (see FT-IR evidence) but it is likely to be due to a smaller particle size in the EV sample. Indeed it is well known that decomposition occurs at lower temperatures in samples of smaller particle size and, on the other hand, the XRD evidence on the EV sample (broad diffraction effects) confirms that it is characterised by small size particles. This fact can explain the differences between these samples and it is likely that they represent one and the same polymorphic form (form E);
- The M and E samples show the same IR spectrum but different XRD patterns. Therefore they show differences in their crystal lattices rather than in molecular geometry. Such structural difference does not appreciably affect the melting enthalpy but it is reflected in a different thermal stability. Indeed the TG/FT-IR experiments indicate that the M sample has the highest thermal stability as can be seen from the fact that it is the only form that evaporates as it is. It can be concluded that M represents a new polymorphic form (form M).

Last we note that our findings, particularly the thermal and IR results, rule out the presence of any solvate.

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