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Physical characterisation of solid forms of cyclopenthiazide

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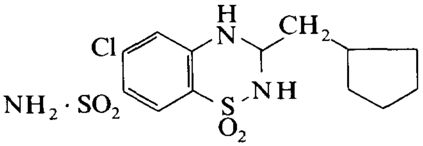
Summary

Three polymorphic forms of cyclopenthiazide were prepared and characterised by DSC, thermomicroscopy, X-ray powder diffraction, scanning electron photomicrographs, IR, solid-state ^{13}C-NMR, solution calorimetry, elementary analysis, intrinsic dissolution, powder dissolution and solubility determinations. Form II has the highest solubility in both 0.01 M HCl and 0.01 M NaOH. It was shown to be metastable in these solvents. When heated continuously, form III melts at 181°C and then recrystallises to form I. Form III is the least soluble in water, 0.01 M HCl and 0.01 M NaOH. Form I has an intermediate solubility in these three solvents and is enantiotropic.

Introduction

The presence of two polymorphic forms for cyclopenthiazide [6-chloro-3-cyclopentylmethyl-3,4-dihydro-2H-1,2,4-benzothiadiazine-7-sulfonamide 1,1-dioxide], a diuretic was reported by Kuhnert-Brandstätter (1971). The melting points found with thermomicroscopy were 238 and 230–236°C for forms I and II, respectively. No other mention of polymorphism of cyclopenthiazide could be found in the literature.



This report deals with the preparation and physical properties of three polymorphic forms of cyclopenthiazide. The three forms were characterised by differential scanning calorimetry, thermomicroscopy, X-ray powder diffraction, scanning electron photomicrographs, infrared spectrometry, solid-state ^{13}C-NMR spectrometry, so-

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lution calorimetry, intrinsic dissolution rates, powder dissolution and solubility determinations.

Materials and Methods

Preparation of crystal forms

Polymorph I was prepared by adding 7.0 g of cyclopenthiazide to a mixture of 20 ml ethanol, 100 ml *n*-heptane and 60 ml of methanol. The mixture was stirred under reflux on a thermal magnetic stirrer until the cyclopenthiazide was dissolved. The hot solution was placed at 20 ° C in a dark room for 4 days. The resulting crystals were filtered off and dried under 600 mm vacuum at 50 ° C for 24 h.

Polymorph II was prepared by dissolving 5.0 g of cyclopenthiazide in 100 ml ethanol. To this solution 20 ml water was added and the mixture heated to 70 ° C. The ethanol was then evaporated with a rotary evaporator. The resulting crystals were filtered off (sintered glass filter) and dried under 600 mm vacuum at 50 ° C for 24 h.

Polymorph III was prepared by adding 5.0 g of cyclopenthiazide to a boiling mixture containing 85 ml of ethanol and 100 ml of water. The mixture was stirred on a thermal magnetic stirrer under reflux until all of the cyclopenthiazide was dissolved. The solution was filtered through a sintered glass filter into a container residing in ice. The resulting solution was then placed in a freezer at 0 ° C for 14 days after which the crystals were filtered off and dried under 600 mm vacuum at 50 ° C for 24 h.

DSC analysis

The thermograms of the different crystal forms were recorded on a Du Pont® 910 DSC System equipped with a Du Pont® Series 99 Thermal Analyzer programmer. A Hewlett-Packard® X-Y recorder was used. The instrument was calibrated with an indium standard. The thermal behaviour was studied by heating 5 mg samples at a heating rate of 5 ° C min^{-1} in a crimped aluminium pan with a crimped empty pan as reference. The study was conducted from 120 to 260 ° C under nitrogen purge.

For the determination of the heats of fusion a Perkin Elmer® DSC-4 differential scanning calorimeter was used.

Thermomicroscopy

The thermal behaviour of the different forms was studied with a Leitz® Laborlux K thermomicroscope equipped with a Leitz® model 350 heating block and a Goerz® 1200 thermocouple. The apparatus was calibrated with analytical grade acetanilide (melting point 115 ° C). The heating rate was 5 ° C min^{-1}.

X-ray powder diffraction

Powder diffraction patterns of the different forms were obtained by using a Philips® PW 1050/70 diffractometer system, CuK_α radiation (λ 1.5418 Å) and a scan speed of 0.5 ° 2θ min^{-1}. The samples were lightly ground and packed into the aluminium sample container.

Scanning electron photomicrographs (SEM)

Photomicrographs were obtained by using the Cambridge® Steroscan 250 scanning electron microscope. The samples were coated under reduced pressure with carbon (Emscope® TB 500 sputter coater) before being coated with a thin gold-platinum film (Eiko Engineering Ion Coater IB-2).

IR spectrometry

Infrared spectra were recorded as potassium bromide discs (preliminary results showed no change during preparation of the discs). The apparatus used was a Nicolet® 5DX FT infrared spectrophotometer, connected to a Nicolet® 5DX mini processor and a Hewlett-Packard® 7470 recorder.

Solid-state ^{13}C-NMR spectrometry

The solid-state ^{13}C-NMR spectral data of the three polymorphs were obtained on a Brucker® AM300 spectrometer using the combined techniques of cross-polarisation and magic angle spinning (CP-MAS) with a 1 ms mixing time, 0.9 s acquisition time and a recycle delay of 3 s. The actual reference compound was adamantly run separately prior to running the samples and using the conversion factor $\delta(\text{CH}_3)_2\delta\text{i} = \delta(\text{adamantance CH}_2) + 37.8$.

NMR solution spectroscopy

(500 MHz, $DMSO_6$) was used to determine that the polymorphs were chemically identical.

Solution calorimetry

The heats of solution were determined according to the method of Winnike et al. (1988) in 95% ethanol. A Tronac® model 450 isoperibol calorimeter was used for determining heat of solution. The bath temperature of 25.000 °C was checked with an NBS-certified thermometer. The temperature was monitored with a Hart® Microtherm model 1006 digital thermometer which gave a constant nominal reading of $25.000 \pm 0.0004\,^{\circ}\mathrm{C}$. The stirring rate was 900 rpm. The heat capacity was measured before and after the determinations. The apparatus was calibrated with an NBS-reference standard (potassium chloride SRM 1655) which was dried at 800 K beforehand. Ampoules developed by Winnike et al. (1988) were used for the samples. The calorimeter was equipped with an Apple® II personal computer through an analog-digital converter and a signal amplifier.

Elementary analysis

Elementary analysis was used to confirm the chemical identity of the polymorphs and to ensure that form II was indeed a polymorph and not a pseudopolymorph.

Intrinsic dissolution rates

The intrinsic dissolution rates of the crystals were determined at 37 °C in 200 ml of a 10% v/v solution of ethanol in water as dissolution medium. Each determination was repeated five times. The method was carried out according to the method of Singh et al. (1968) and Simonelli et al. (1969). The crystals were compressed into 13 mm discs in an evacuable die at 705 MN m^{-1} (10 ton). The dissolution medium was stirred at 150 rpm and the cyclopenthiazide dissolved was determined spectrophotometrically (Shimadzu® model UV 2100) at 225.3 nm.

Powder dissolution

Powder dissolution rates were determined by a modification of the method described by Lötter et al. (1983). Each determination was repeated five times. The dissolution medium was 500 ml distilled water at 37 °C. Samples were passed through a 50 μm sieve and 50 mg of each form was then suspended in 50 ml water on an ultrasonic bath. 1 ml of this suspension was added to the dissolution bath and stirred at 100 rpm. At each time interval, 5 ml samples were withdrawn and the contents determined spectrophotometrically at 225.3 nm.

Solubility determinations

A sufficient quantity of crystals of each polymorph to effect saturation was placed in 5 ml ampoules containing water, 50% v/v ethanol in water, 0.01 M hydrochloric acid or 0.01 M sodium hydroxide. Duplicate ampoules of each form in each solvent were sealed and stirred at 30 rpm for 7 days with a Heidolph® R2R2000 stirrer

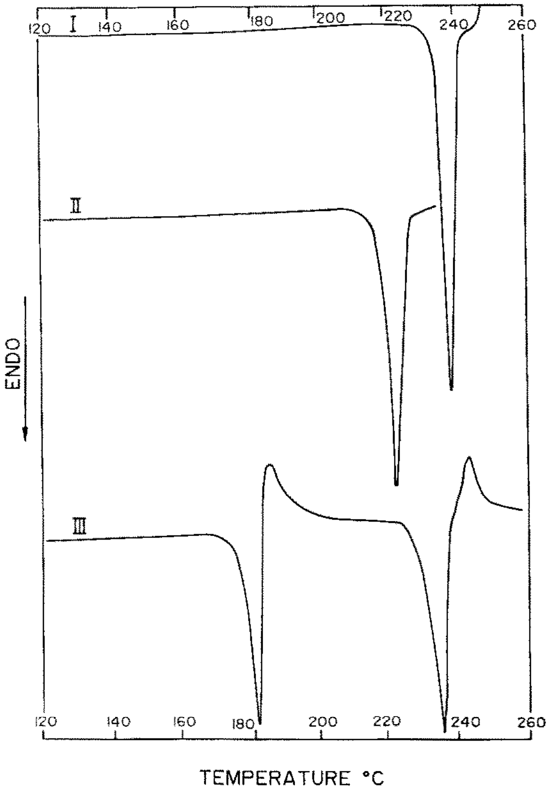

Fig. 1. DSC thermograms of the three polymorphs of cyclopenthiazide with melting points as follows: form I, 238 °C; form II, 225 °C; form III, 181 and 235 °C.

connected to a stirring device. The temperature was maintained at 37 ° C with a Julabo® PC thermostat. After the 7 days the ampoules were opened, the content filtered and the concentration determined spectrophotometrically at 224.9 nm for 0.01 M HCl, 220.1 nm for 0.01 M NaOH and 225 nm for 50% ethanol in water.

Interconversion of the forms in contact with liquid at 37 ° C

The stability of the forms at 37 ° C in contact with the liquids of the solubility study was checked by subjecting the filtered off part of each solvent to DSC.

Results and Discussion

Three polymorphic forms of cyclopenthiazide were prepared and characterised by using DSC, thermomicroscopy, X-ray powder diffraction, scanning electron microscopy, IR, solid-state ^{13}C-NMR, solution calorimetry, intrinsic dissolution rates, powder dissolution rates and solubility determinations.

DSC

The melting points of the three forms derived from the thermograms (see Fig. 1) were found to be as follows: form I, 238 ° C; form II, 225 ° C; form III, 181 and 235 ° C.

In order to determine the heats of fusion these determinations were repeated on the Perkin Elmer® apparatus. The results of melting point and heats of fusion are given in Table 1.

It is clear that differences in heats of fusion do exist and that melting points measured with the two apparatuses are in good agreement with each other.

TABLE 1

Melting points and heats of fusion for three polymorphs of cyclopenthiazide

Polymorph	Melting point (° C)	Heat of fusion (J/g)
I	239.33	277.67
II	223.03	259.09
III	181.87 and 233.48	164.35

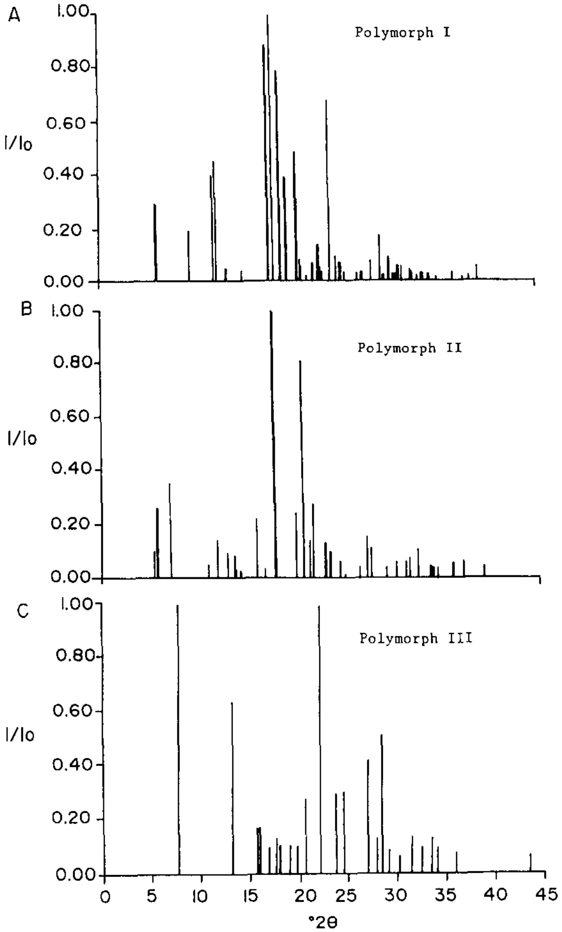

Fig. 2. X-ray powder diffraction. The results of I/I_0 plotted against ° 2θ as obtained from the powder diffractograms.

Thermomicroscopy

Forms I and II showed only a single melting point at 238 and 225 ° C, respectively. However, it was clear that at 181° C form III melts and then recrystallisation to form I takes place which then melts at 238 ° C. This conversion was confirmed by recording a thermogram of form III with DSC and then heating it in an oven at 180 ° C for 10 min. The DSC thermogram thereafter showed only the thermogram of form I.

X-ray powder diffraction

The results of I/I_0 plotted against °2θ are compared in Fig. 2. It is clear that major differences between the forms exist.

Scanning electron photomicrographs

The SEM photomicrographs are given in Figs 3–5. The differences can clearly be seen.

Infrared spectrometry

The FTIR spectra of the three forms are compared in Fig. 6. The spectrum of form I is given on the bottom part in each instance for comparison purposes. It is clear that differences between the spectra are evident at the wavelengths indicated by the arrows.

Solid-state ^{13}C-NMR

The spectra of the three forms are compared in Fig. 7. Major differences are evident from the spectra.

Solution calorimetry

The heats of solution of the three polymorphs are given in Table 2.

No statistical significant difference exists between forms I and II. This may have been due to the presence of moisture in the sample of form II as the sample was not dried before the determination. In the case of form III there is a major difference from forms I and II.

TGA and DSC data indicated that form II is a true polymorph and not a pseudopolymorph. NMR solution spectroscopy (500 MHz, $DMSO_6$) and elementary analysis performed on all three polymorphs confirmed that they were chemically identical ($C_{13}H_{18}ClN_3O_4S_2$).

Intrinsic dissolution rates

The results of the intrinsic dissolution rates are given in Fig. 8.

The least-square fit for a straight line through the dissolution points was calculated for each

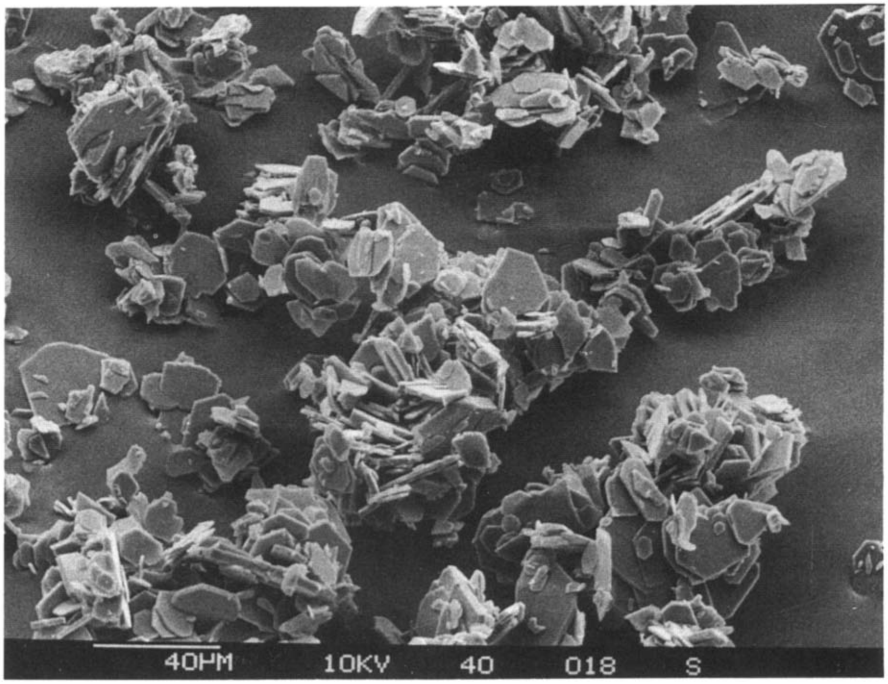

Fig. 3. Scanning electron photomicrograph of the crystals of form I.

TABLE 2

Heat of solution for polymorph I, II and III

Polymorph	Heat of solution (cal mol^{-1})
I	1412 ± 258
II	1467 ± 183
III	3584 ± 129

dissolution curve. The gradient was taken as the intrinsic dissolution rate.

The intrinsic dissolution rates were compared using the Student-Newman-Keuls Multiple Range Test. The intrinsic dissolution rate of polymorph II was significantly higher (95% confidence level) than the rates of any of the other polymorphic forms. The dissolution rate of polymorphic forms I and III did not differ significantly.

Thus the metastable form II showed the best solubility, followed by form I and III.

Powder dissolution

The mean particle size of the three forms as was determined by Coulter Counter was as follows: form I, 24.16 μm; form II, 25.21 μm; form III, 20.85 μm.

As was the case with intrinsic dissolution form II gave the best dissolution followed by form I. Form III had the slowest dissolution initially.

Solubility study

The results are given in Table 3. Form II is best soluble in all solvents followed by form I and then form III which is the least soluble in all solvents.

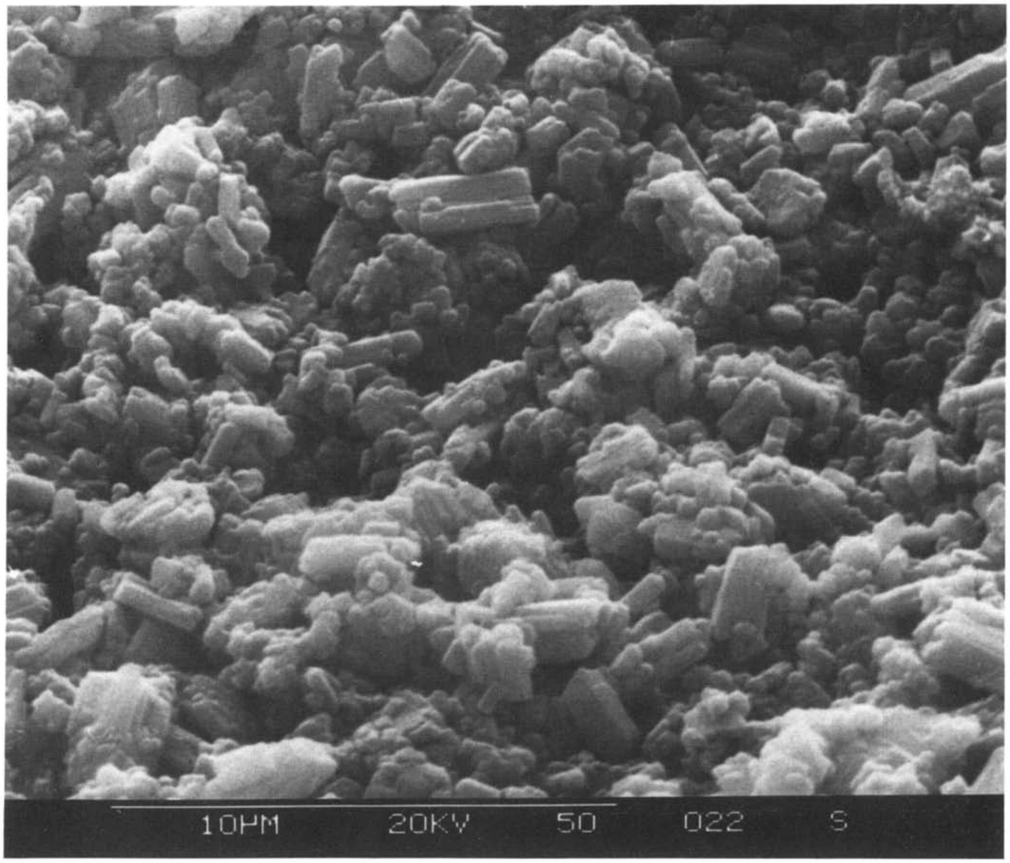

Fig. 4. Scanning electron photomicrograph of the crystals of form II.

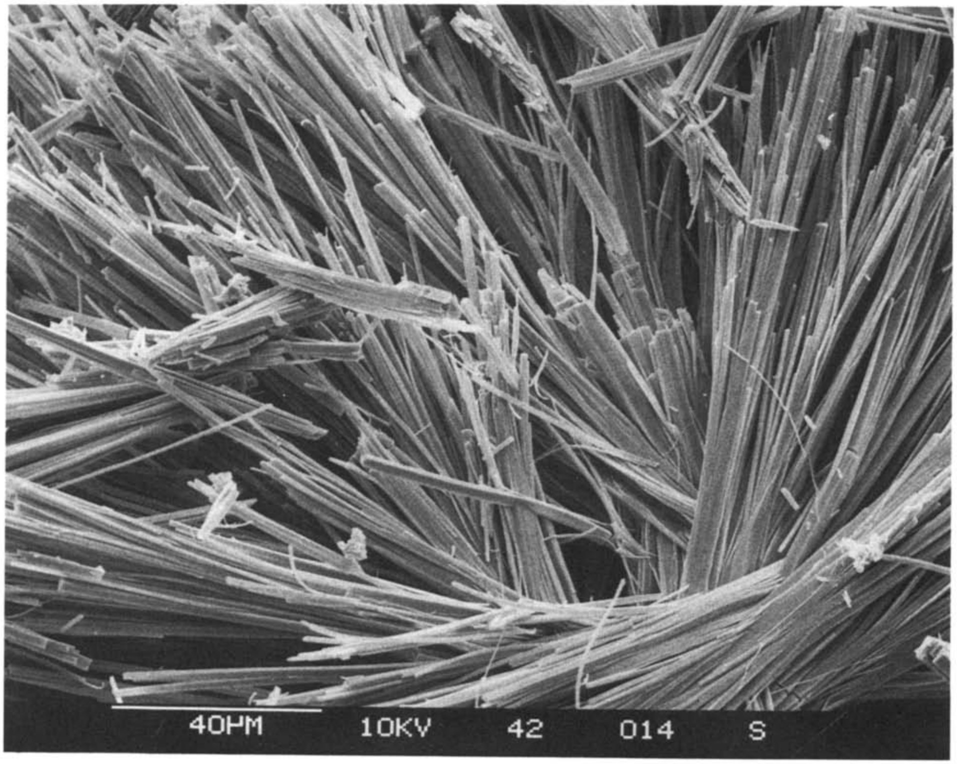

Fig. 5. Scanning electron photomicrograph of the crystals of form III.

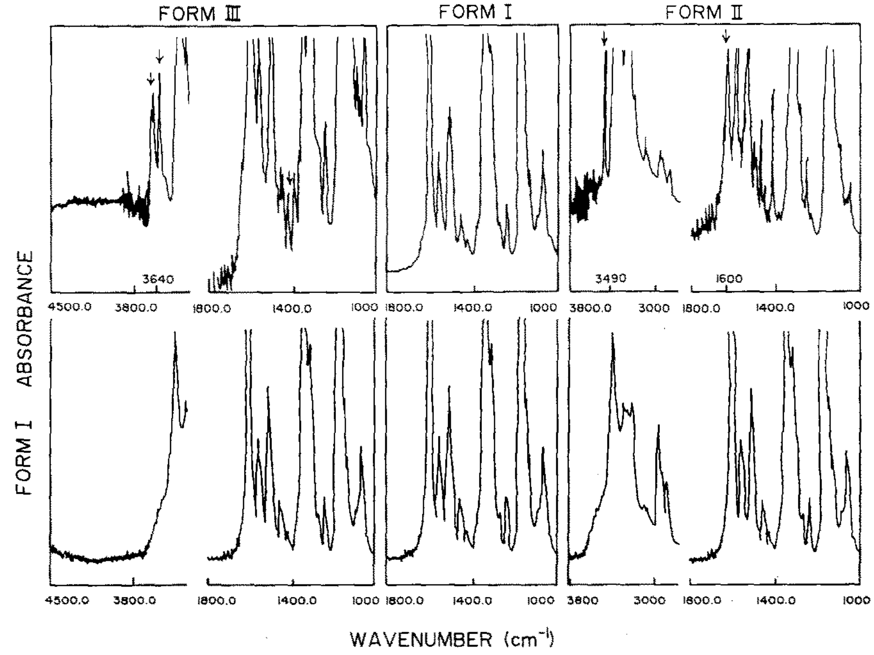

Fig. 6. The compared FTIR spectra of the three polymorphs of cyclopenthiazide. Differences in spectra are indicated by arrows.

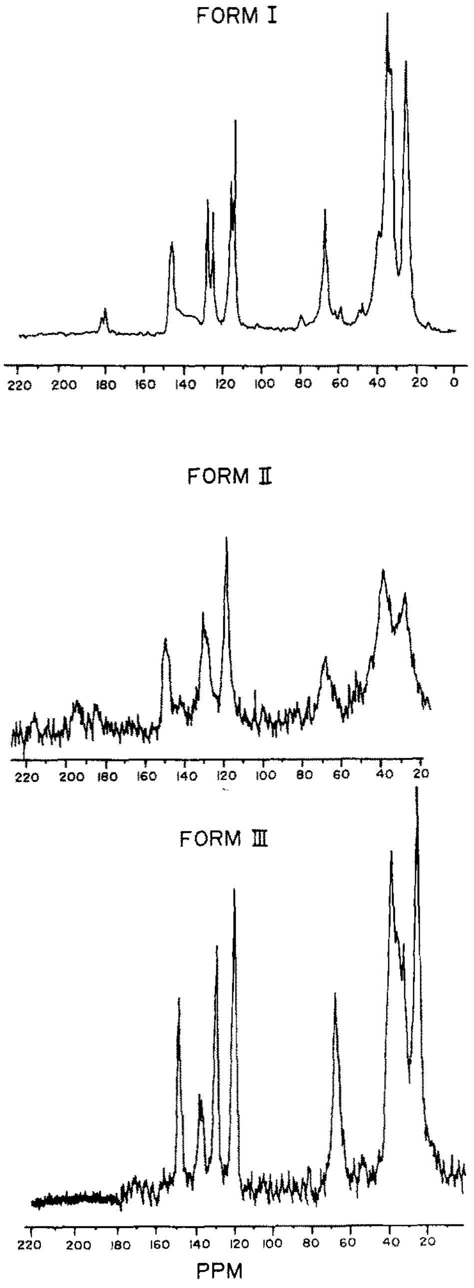

Fig. 7. The compared solid-state ^{13}C-NMR spectra of the three polymorphs of cyclopenthiazide.

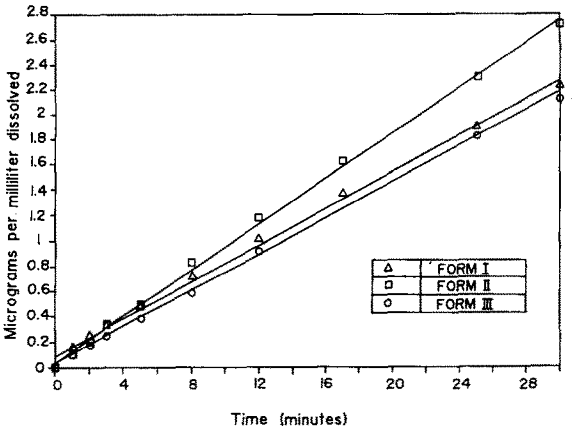

Fig. 8. Intrinsic dissolution rates of the three polymorphs of cyclopenthiazide.

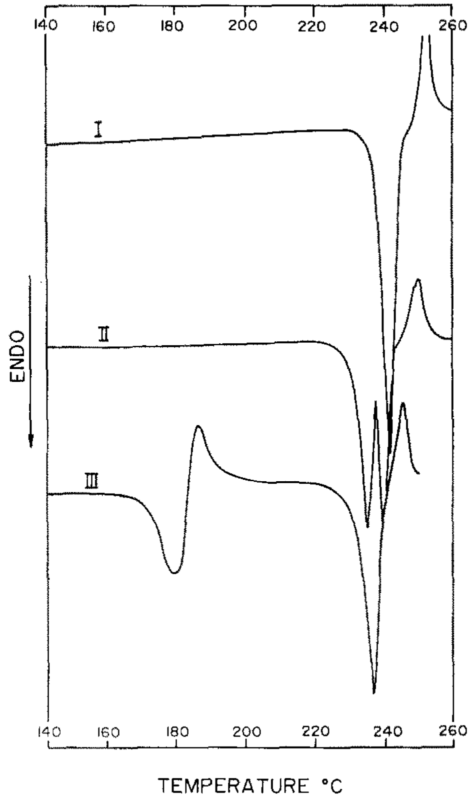

Fig. 9. Interconversion of polymorphic forms in 0.01 M NaOH.

Interconversion of polymorphic forms in liquid

The DSC thermograms taken of the residue after the solubility study showed that form II converts to form I in both 0.01 M HCl and 0.01 M NaOH but not in water or 50% ethanol in water. The conversion as could be seen from the DSC thermogram of form II from the solubility in 0.01 M NaOH is given in Fig. 9 as an example.

TABLE 3

Solubility of the three polymorphs in water, 50% ethanol in water, 0.01 M HCl and 0.01 M NaOH

Polymorph	Solubility (in μg ml^{-1})			
	Water	50% ethanol in water	0.01 M HCl	0.01 M NaOH
I	34.70	200.74	23.90	133.07
II	61.80	496.13	62.63	306.33
III	17.15	132.40	13.16	129.16

All solubility data thus suggest that polymorph III is thermodynamically the most stable of the three polymorphs. Forms III and I did not show any conversion in all the tested solvents.

The solubility of form I lies between that of form II and III in all tested solvents. To determine whether form I is enantiotropic or monotropic a solubility study was repeated where seed crystals of form III were mixed with form I crystals before the study. After 7 days the solubility was found to be 36.92 μg/ml which was not significantly higher than that found during the actual solubility study, i.e. 34.70 μg/ml. When conversion to form III was present the solubility should have lowered to 17 μg/ml. It can thus be stated that polymorph I is monotropic.

Conclusions

The physical characterisation of three polymorphs of cyclopenthiazide was carried out. Differences (not significant in all cases) between the forms were found with all methods.

All solubility data indicate that polymorph II is the best soluble followed by I and then III which is the least soluble. These differences were small during intrinsic and powder dissolution. Significant differences were, however, found during the solubility study. Form II with a good solubility would probably be the best form for tablet manufacture. However, it is metastable and difficult to prepare. The poor solubility of form III precludes its use in solid dosage forms.

During this study commercial samples were also scanned for the presence of polymorphs. One sample containing polymorph III had the lowest solubility of all the tested samples. When polymorph III is thus detected in a raw material it is suggested that recrystallisation to form I should be performed.

Polymorph III crystallises from water/ethanol mixtures. The presence of water during synthesis of cyclopenthiazide or during the manufacture of tablets could lead to conversion to polymorph III with a subsequent lower solubility.

Polymorph I is the form of choice for the production of solid dosage forms. Its solubility is about 50% better in all solvents tested than polymorph III. The metastable form II is about 50% more soluble than polymorph I in all solvents tested.

When solubility data and the stability at 37 °C in liquids are taken into account, consideration should be given to change the naming of form III to form I.

Acknowledgement

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