Application of differential scanning calorimetry to the study of sulphathiazole crystal forms

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Methods of preparation and characterization of two crystal forms of sulphathiazole using differential scanning calorimetry and infrared spectroscopy are described. Heats of transition and fusion calculated from thermal analysis are 1778 ± 68 Cal mol^{-1} and 6615 ± 235 Cal mol^{-1} respectively. The transition temperature depends on the rate of heating and the sample source. Interconversion of the crystal forms under different conditions such as heating and suspension in water is described. An assay procedure based on the area of the thermal transition peak of Form I was devised and results of analysis of synthetic mixtures of Forms I and II are given.

The existence of sulphathiazole in different polymorphic forms was first reported by Grove & Keenan (1941) who described methods of preparation and characterization of two crystal forms. Later, Miyazaki (1947) added a third form which melted at 173–175° without polymorphic transformation. Mesley & Houghton (1967) reported the existence of three crystalline forms, which they correlated with Miyazaki's forms, and added also a fourth amorphous modification. Solubility, dissolution rate and thermal behaviour of sulphathiazole have been studied by Brandstaetter-Kuhnert & Martinek (1965); Milosovich (1964); Inoue & Saito (1961) and Guillory (1967). In most of these publications, methods of preparation and characterization of only two crystalline modifications are described.

A re-examination of this problem using differential scanning calorimetry is described below. The number of forms is of direct relevance to the B.P.C. infrared identification test and the preparation of stable aqueous suspensions of sulphathiazole.

EXPERIMENTAL AND RESULTS

Apparatus

A Perkin-Elmer DSC-1 differential scanning calorimeter fitted with effluent analyser was used. Samples of pure indium were used to calibrate the instrument. Dry nitrogen at 30 ml/min was used as carrier gas. Various rates of heating were employed ranging from 0·5° to 64°/min. Samples usually varied from 1–10 mg and were weighed with an electromicrobalance (EMB-1 Research and Industrial Instruments Co.).

Infrared spectra of Nujol mulls were recorded on a Unicam SP200 double beam spectrometer fitted with a sodium chloride prism (Fig. 1). Wavelength calibration with a polystyrene film showed an accuracy of ± 3 wavenumbers.

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Material

Two batches of sulphathiazole were used; a B.P.C. authentic specimen and a micro-milled material (May and Baker Ltd.) of B.P.C. quality.   Both were of Form I.

Preparation of the crystal forms

Form I was prepared by crystallization of sulphathiazole from either boiling water, acetone-chloroform mixture, dilute ammonia solution or 95% aqueous ethanol. The crystals were dried by passing a current of air through them on the filter funnel. Crystal crops were stored in closed containers in a cool place.   Different solvents produced crystals of varying shapes and sizes but all were placed in one category since they possessed similar properties (see below).

Form II was obtained by crystallization from s-butanol or n-amyl alcohol; it was also prepared by heating Form I to 180°.   Neither infrared spectroscopy nor differential scanning calorimetry showed the presence of solvents in any of the samples.

Characterization of the crystal forms

The infrared spectra of Forms I and II in Nujol mulls (Fig. 1) showed differences which correspond to the Forms I and II described by Guillory (1967).
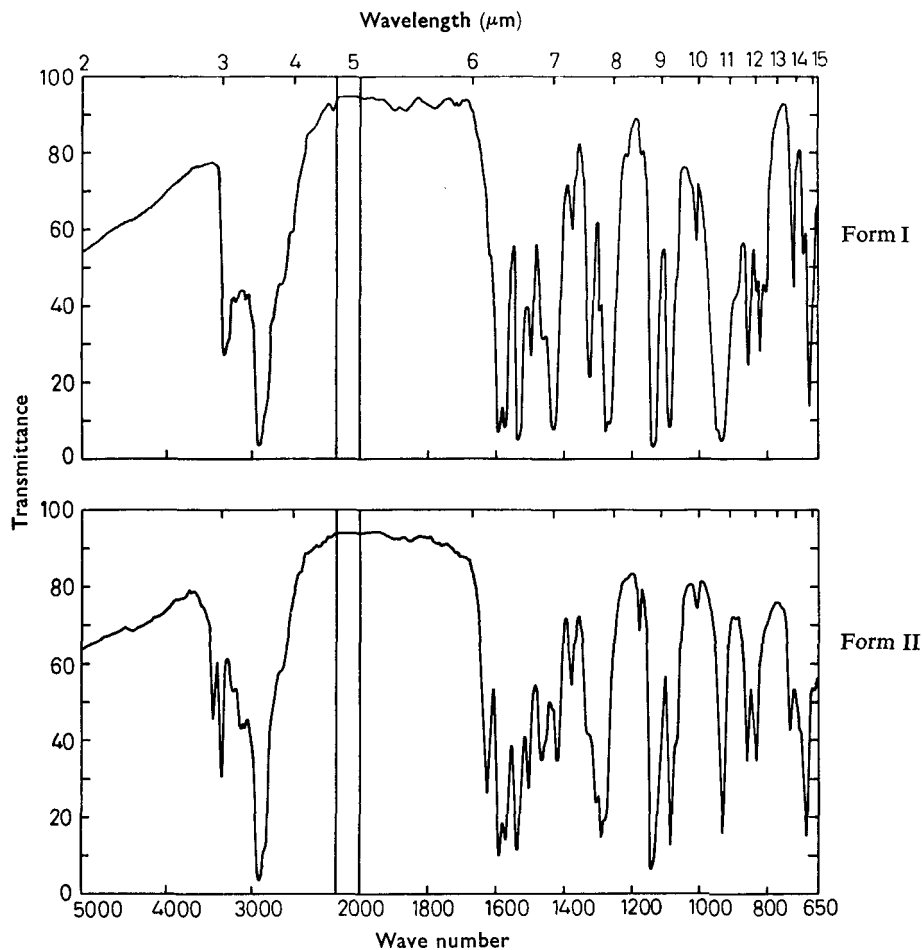

FIG. 1.   Infrared spectra of sulphathiazole crystal forms in Nujol mulls.

While with differential scanning calorimetry Form II showed only one endothermic transition corresponding to melting at *ca* 205°, Form I showed two endothermic transitions. The first corresponds to polymorphic transformation of Form I to II and the second to melting. In no case has melting been observed at temperatures below 200° (cf. Miyazaki, 1947). The temperature of both transitions, but especially the first, varied with the rate of heating, shape and size of crystals and crystals prepared under different conditions for (different solvents and different rates of crystallization—see Table 1).

Table 1. *Effect of sample source and rate of heating on transition temperatures of sulphathiazole*

Sample	Temp. at peak max of 1st transition (°C)		Temp. at peak max of 2nd transition (°C)	
	16°/min	32°/min	16°/min	32°/min
B.P.C. specimen	174	—	207	—
Micromilled preparation ..	174	180	207	213
Form I—boiling water ..	158·5	—	207	—
Form I—acetone/chloroform ..	163	169	208	212
Form I—dil. ammonia ..	176	181	208	214
Form I—95% ethanol	165, 176·5	175, 181	206	213
Form I—susp. of II in water ..	139	—	206	—
Form II—s-butanol	—	—	—	213
Form II—amyl alcohol ..	—	—	200·5	206

Guillory (1967) and Grove & Keenan (1941) have previously drawn attention to the effect of rate of heating on the transition temperature. However, the first transition was observed for all samples of Form I in the temperature range 140–180°. Fig. 2 shows typical behaviour of some forms and mixtures of them. In some, e.g. Form I prepared by crystallization from 95% aqueous ethanol, two transitions have been observed corresponding to the transformation to Form II. Two species of Form I are
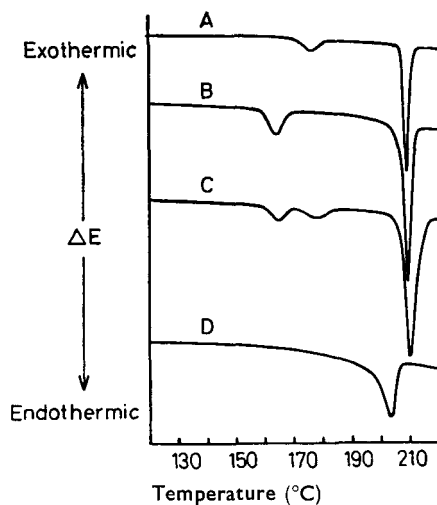

Fig. 2. Thermal analysis curves of sulphathiazole crystal forms ($\triangle$E is proportional to milli-calories/s rate of heating = 16°/min). A. Form I. B.P.C. B. Form I, crystallized from acetone–chloroform. C. Mixture of Form I crystallized from acetone–chloroform and Form I crystallized from dilute ammonia. D. Form II, crystallized from amyl alcohol.

apparently produced in different proportions from this solvent. No effluent vapour was detected for any of the samples studied. Rate of heating had no effect on peak areas in the range of 8–32°/min, and consequently heats of transition are constant. Different species of Form I have the same heat of transition.

The heats of transition and fusion were calculated from peak areas (measured by planimeter), using the calibration curve of Carless, Moustafa & Rapson (1966). These were 1778 ± 68 Cal mol^{-1} ($P = 0.95$) for the first transition in the authentic B.P.C. specimen and 6615 ± 235 Cal/mol^{-1} ($P = 0.95$) for melting of Form I crystallized from 95% aqueous ethanol.

Interconversion of the crystal forms

(a) *Crystallization.* Forms I and II may be interconverted by crystallization from the appropriate solvent (see above). Only two forms were encountered. The change of Form II to I after suspension in water was detected by filtering off the solid which was dried in a current of air, and then examined by infrared spectroscopy and differential scanning calorimetry.

(b) *Heating.* Form I changes to Form II on heating to 180°. This is in agreement with results of previous workers (Milosovich, 1964; Brandstaetter-Kuhnert & Martinek 1965, among others). Although the change was rapid above the transition temperature, prolonged heating below this had no effect on the crystal form. Transformation from Form I to Form II was independent of the rate of heating.

(c) *Suspension in water.* It has been reported (Sanchez, 1962) that the metastable form of sulphathiazole is readily transformed into the stable form in the presence of moisture. In the present study, Form II changed to Form I upon suspension in water—the time necessary to achieve this transformation varied according to the source of the crystals and their size. For Form II prepared by crystallization from s-butanol, this was about two weeks at room temperature.

(d) *Grinding.* Dry grinding of either crystal form by hand or in a vibration mill for periods up to 5 min had no detectable effect on the infrared spectra of the crystals. Vibration milling of Form I crystallized from 95% ethanol resulted in the transition temperature being lowered by about 5°.

Estimation of Form I in mixtures of crystal forms

Since only Form I shows an endothermic transition in the temperature range 125–180°, use has been made of this in the estimation of this form. A calibration curve (Fig. 3) was established relating the peak-area of this transition to the sample weight using the authentic B.P.C. material. Using this curve, a series of mixtures of Forms I and II and of Form I from different sources were analysed. In the latter case the total area of any peaks in the temperature range 125–180° was measured. Results of these assays are shown in Table 2.

DISCUSSION

Few papers have appeared in the literature in which three or more forms have been reported (Miyazaki, 1947; Mesley & Houghton, 1967). In the present investigation only two crystal forms were identified, none of which melts at 175° as suggested by these authors.

Reported transition temperatures for Form I to Form II show wide variation. Using solubility data, Milosovich (1964), reported a low transition temperature of
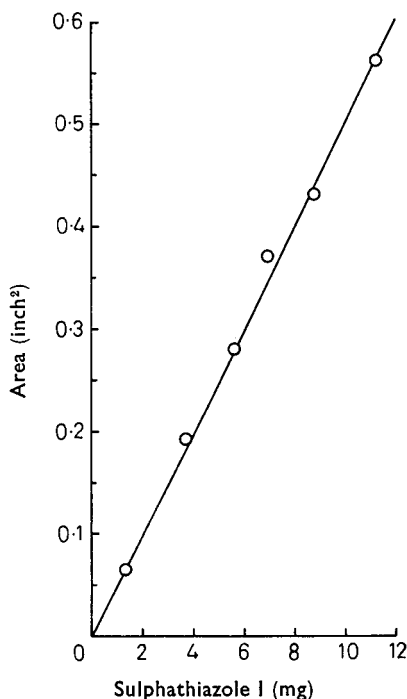
FIG. 3. Calibration curve for the first thermal transition in sulphathiazole Form I. (Rate of heating = 16°/min, range 4, chart speed = 0·75 inch/min).

Table 2. *Determination of Form I in mixtures of other crystal forms, by differential scanning calorimetry*

Mixture		Form I found mg	Error
Form I (BPC authentic specimen) mg	Other materials mg		
2·174	5·948 Form II	1·83	—16%
5·572	5·914 Form II	4·67	—16%
8·814	1·940 Form II	8·12	—8%
8·825	3·335 Form II	7·90	—10%
5·260	3·396 Form II	5·07	—4%
2·302	5·214 Form II	2·01	—13%
8·546	4·550 *Form I	11·97	—9%
4·560	6·226 †Form I	10·55	—2%

N.B. *Form I—crystallized from dilute ammonia.
† Form I—crystallized from acetone chloroform mixture.
All Form II was crystallized from n-amyl alcohol.

94·5° ± 2·7°, whilst using differential thermal analysis transition temperatures as high as 161° have been reported by Guillory (1967). Intermediate values are given by Inoue & Saito (1961). The present investigation has shown variation between different samples of Form I. The rate of heating, method of preparation, and particle size, affect the transition temperature (Table 1). The shape and size of the crystals will affect the thermal conductivity and hence transition temperature. However, further investigation is necessary to determine whether Form II crystallized from amyl alcohol differs from the other forms, or whether its lower melting point is due solely to

particle size effects.   In spite of variations in transition temperatures, the constancy of the energy uptake appears to offer a method for the estimation of Form I in a mixture of the two forms.

Different methods of preparation of sulphathiazole crystal forms has led to some confusion.   Mesley & Houghton (1967) obtained Form C, which corresponded to Miyazaki's (1947) Form α′, by crystallization from dilute ammonia solution. Mesley & Houghton also reported that the infrared spectrum of this form was in fact very similar to that of Form A.   In the present study, recrystallization from dilute ammonia produced Form I which should be identical with Mesley & Houghton's Forms A or C. In spite of the fact that this had a somewhat different transition temperature from other varieties of Form I, no melting was observed at 175° when it was heated on the DSC-1 or in a Gallenkamp melting point apparatus [cf. Miyazaki's (1947 form α′, m.p. 175°].
This form therefore appears to be another variety of Form I since it has the same infrared spectrum and heat of transition and it is not justifiable to call it a new form. Grove & Keenan (1941) Milosovich (1964) and Brandstaetter-Kuhnert & Martinek (1965) all quoted a melting point of 175° for Form I which could be due to heating the sample at a rapid rate when melting occurs at 175° without polymorphic transition, as noted by Guillory (1967).   Variation of the transition temperature of I and II could be interpreted on the basis of the factors discussed earlier.   Recrystallization from ethanol produced Form I, contrary to the findings of Mesley & Houghton (1967) who reported that it produced substantially form B.

The heat of transition calculated in the present investigation (1778 ± 68 Cal mol^{-1}) shows good agreement with the value of 1744 Cal mol^{-1} calculated by Milosovich (1964) from solubility measurements of the two crystal forms. However, Guillory (1967) using differential thermal analysis reported a heat of transition of 1420 ± 40 Cal mol^{-1}.   The heat of fusion found in the present work (6615 ± 235 Cal mol^{-1}) is also slightly but consistently higher than that reported by Guillory (1967): viz. 5960 ± 210 Cal mol^{-1}.

The use of differential scanning calorimetry for the estimation of Form I of sulpha-thiazole has been shown to be feasible though not highly accurate (Table 2).   Experi-mental values are always less than the theoretical, and this may be due to the mixing of samples of different particle shapes and sizes.   The heterogeneity of the sample mixture leads to a slight decrease in the area of the transition peak.   This effect is non-specific and has been observed for mixtures of Forms I and II and also for mixtures of samples of Form I from different sources (see Table 2).   No reasonable correction factor relating the transition peak area at the fusion peak area could be found because admixture of a second sample was found to affect both peaks to different degrees. The errors were smaller when one variety of Form I was the major component in a mixture, interference being in this case at a minimum.   This method however, is useful in giving an approximate measure of the proportion of various forms of mixture, Form II being calculated by difference from the total.   This assay procedure is obviously useful when rates of transformation in aqueous suspensions are being studied.

Previous suggestions (Carless & Foster, 1966) that the B.P.C. authentic specimen was a mixture of two forms must be modified since the present study suggests that it consists mainly of Form I alone.   Thermal behaviour and infrared spectrum are typical of this form.   For the purposes of the identification test, it is suggested that any sample of sulphathiazole should be heated to a temperature of 180°, then its infrared spectrum determined in Nujol mull.   The infrared spectrum of Form II should then

be reproduced in all cases. This is a simpler procedure than the alternative of Mesley & Houghton (1967) which involves recrystallization from n-propanol.

Accelerated crystal growth in aqueous suspensions of sulphathiazole by temperature cycling has been studied by Carless & Foster (1966). No polymorphic changes were detected in these systems which consisted of Form I suspensions (Foster, personal communication). It would be of interest to study the particle size distribution and polymorphic changes of Form II in aqueous suspension as this may be analogous to the cortisone acetate suspensions where crystal growth is initiated mainly by polymorphic transformation (Moustafa, 1967). If this was so then the use of Form I of sulphathiazole would be necessary for the preparation of stable aqueous suspensions.

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