

# RESEARCH PAPERS

## OPTICAL CRYSTALLOGRAPHIC PROPERTIES OF SOME LOCAL ANÆSTHETICS

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THE observation of optical crystallographic properties of crystalline compounds is a good micro-identification method, which is gaining ground in pharmaceutical analysis. Recent chemical literature includes several publications reporting the optical crystallographic data of commonly used drugs within important pharmacological groups, as for example barbiturates,<sup>1</sup> sulphonamides,<sup>2,3</sup> sympathomimetic amines,<sup>4,5</sup> and anti-histaminic drugs.<sup>6,7,8</sup> The purpose of the present paper is to report the distinctive optical characters of the hydrochlorides of some local anæsthetics. Hucknall and Turfitt<sup>9</sup> have described a simple isolation technique by which the hydrochlorides of the local anæsthetics may be obtained in a high degree of purity from biological materials or medicinal preparations. Also, since the commercially available salt of these bases, with few exceptions, is the hydrochloride, the optical data of the hydrochlorides should be useful in most cases where the identification of a local anæsthetic is required on a micro scale.

The principal refractive indices of the hydrochlorides of procaine, tetracaine (amethocaine), phenacaine and *l*-cocaine are listed in a determinative table published by Keenan<sup>10</sup> and reprinted in U.S.P. XIV, pp. 747 to 749. Keenan<sup>11</sup> has also reported the optical crystallographic properties of the precipitates obtained from solutions of procaine and butacaine on addition of solid potassium iodide. van Zijp<sup>12</sup> has described a method for the differentiation of procaine hydrochloride and *l*-cocaine hydrochloride by observations in the polarising microscope without determination of the refractive indices. In the present paper some optical data are reported for the hydrochlorides of butacaine, cinchocaine, larocaine and tutocaine. Butacaine hydrochloride proved to be polymorphic. The polymorphism of tetracaine hydrochloride has been observed in melting point determinations by Doser.<sup>13</sup> The refractive indices of tetracaine hydrochloride are recorded in U.S.P. XIV without reference to the polymorphism of the substance. The present paper reports refractive indices of two pure phases of crystalline tetracaine hydrochloride, which are both frequently present in commercial samples. The optical crystallographic identification of *l*-cocaine hydrochloride should be of particular interest since this substance decomposes on melting<sup>14</sup> and its melting point is variously reported in the literature.<sup>9</sup> Commercial or recrystallised samples of this substance often consist of irregular fragments. For identification purposes it is therefore advantageous to

examine in the polarising microscope the well developed crystalline sublimate obtained at 165° to 170° C. The optical characters of this sublimate are described below.

The principal refractive indices ( $\pm 0.002$ ) of the compounds which have been examined in the present work, are listed in Table I according to the ascending value of the lower index. The optical crystallographic properties have been determined as completely as the available equipment and the common orientations of the compound in question would allow. The refractive indices of the crystals were determined (at 20° C.  $\pm 1^\circ$ ) by the immersion method for yellow, filtered light (glass filter). The refractive indices of the immersion liquids were measured (at the same temperature) by means of a Spencer Abbé-type refractometer. It should be mentioned that the substances examined in the present work are rather soluble in some of the pure organic liquids usually employed as immersion liquids.<sup>15</sup> Mixtures of liquid paraffin,  $\alpha$ -bromonaphthalene and methylene iodide do not dissolve the crystals. The principal refractive indices ( $\alpha$ ,  $\beta$ ,  $\gamma$ ) were determined with reference to the orientation of the optic axis, observed on interference figures in convergent polarised light. The higher and the lower index ( $\alpha$  and  $\gamma$ ) were then confirmed by determinations on crushed material, the random orientation of the crystal fragments being insured by moving the cover glass. If a principal refractive index has been determined exclusively by this statistical method, it is noted in the following report. Significant intermediate refractive indices ( $n_1$ ) are reported when they may be useful in the identification of a substance. An intermediate refractive index is of little diagnostic value if the corresponding orientation cannot be ascertained by other observations (e.g., extinction angle).

The melting point determinations were carried out, according to the technique described in detail by Kofler,<sup>14</sup> in a micro apparatus of the Kofler pattern. The microscope was equipped with a polariser and an analyser in order to facilitate the observation of any possible conversion from one crystalline phase to another during the heating of polymorphic substances. Changes of the birefringence of the crystals thus observed during the heating (Table I, Nos. 1, 2 and 8), were always confirmed by the use of immersion liquids.

Polymorphism is not unusual among organic compounds of therapeutic interest,<sup>14,16</sup> and crystallographic data are sometimes reported without account being taken of the existence of two or more crystalline phases, since the lower-melting phases are often not detected when the melting point determination is carried out in a glass capillary. Some advantages derived from combining optical crystallographic methods with observations in the micro melting point apparatus of Kofler are evident from the experimental section.

## EXPERIMENTAL

### *Larocaine*

Hydrochloride of 3-diethylamino-2:2-dimethylpropyl *p*-aminobenzoate.  
*Melting point* of the sample (D.A.B. VI. standard): 197° to 198° C.

Between 160° C. and 185° C. a sublimate is obtained,<sup>14</sup> which has a crystallographic orientation of diagnostic value.

*Observations in parallel polarised light* (crossed Nicols). Crystals with rectangular silhouette and parallel extinction. Rods showing high order interference colours and negative elongation, or thinner plates showing positive elongation. The plate-like form predominates if the crystals have been precipitated with ether from an ethanolic solution.

*Observations in convergent polarised light* (crossed Nicols). The common orientation gives the optic normal interference figure.

*Refractive indices.*  $\alpha = 1.521$  and  $\gamma = 1.742$  are obtainable from the common orientation of the crystals.  $\beta$  could not be measured with reference to any interference figure in convergent polarised light. The rods could be rolled into an orientation (form: needles) from which  $\beta$  could be estimated but not exactly determined because of the instability of this orientation.

*System*, orthorhombic. The crystallographic plane parallel to the optic plane (100) is well developed.

*Distinctive characters of the sublimate.* Triangular plates with two of their edges slightly curved outward.<sup>14</sup> They are lying on the plane parallel to the optic plane, showing the refractive indices  $\alpha$  and  $\gamma$ .  $\gamma$  (Z) is parallel to the cleavage lines often observed.

#### *Cinchocaine Hydrochloride*

Hydrochloride of 2-butyloxycinchonic  $\beta$ -diethylaminoethylamide.

*Melting point* of the sample (B.P. standard): 97° to 100° C.

*Observations in parallel polarised light* (crossed Nicols). Rectangular rods or plates with parallel extinction and positive elongation. Well developed crystals showing this form may be obtained by recrystallisation from benzene or from ethanol-ether.

*Observations in convergent polarised light* (crossed Nicols). The optic normal interference figure is commonly observed.

*Refractive indices.*  $\alpha = 1.521$  and  $\gamma = 1.645$  are obtainable from the common orientation of the crystals.  $\beta$  was not determined for the reason given above.

*System*, orthorhombic. The crystallographic plane parallel to the optic plane is well developed.

#### *Tutocaine*

Hydrochloride of (*dl*)-3-dimethylamino-2:1-dimethylpropyl *p*-aminobenzoate.

*Melting point* of the sample (Bayer): 213° to 215° C. From about 160° C. sublimation occurs. The condensate is mainly liquid, above 190° C. also crystalline.

*Observations in parallel polarised light* (crossed Nicols). Small needles or lamellæ showing negative elongation and parallel (or nearly parallel) extinction. The crystals are too small for reliable observations in convergent polarised light. Recrystallisations did not give crystals of more suitable size for these observations.  $\alpha$  is observed lengthwise on all

clear cut needles showing parallel extinction and crosswise  $\gamma$  is frequently found.

*Refractive indices.*  $\alpha = 1.514$  and  $\gamma = 1.679$  were determined from crushed material and verified on some crystals of the sublimate showing an optic normal interference figure.  $\beta$  was not determined.

*System,* probably orthorhombic.

*Distinctive characters of the sublimate.* The crystalline sublimate consists of needles and lamellæ, often with outward curved edges. Elongation negative.  $\alpha$  is frequently observed. Some of the crystals show the optic normal interference figure ( $\alpha$  and  $\gamma$ ). Determination of the refractive indices from the sublimate may be difficult because of the numerous drops condensed on the cover glass.

### *Butacaine Hydrochloride*

Hydrochloride of 3-di-*n*-butylaminopropyl *p*-aminobenzoate.

*Melting points* of the sample (Abbott laboratories. Analysis: argentometric titration, 100.0 per cent., titration with sodium nitrite, 97.54 per cent.) The highest melting point observed was 151° C.<sup>9</sup> During the heating changes in the birefringence and extinction directions of the crystals were observed at temperatures above 100° C., especially between 110° and 120° C. Some of the crystals were then seen to melt at 131° C. In the drops of liquid substance long needles with parallel or nearly parallel extinction and negative elongation grew rapidly at temperatures between 132° C. and 136° C. These needles were seen to melt at 151° C. together with the rest of the crystals. If the substance was placed on the heating block of the microscope previously heated to a temperature between 110° and 120° C., all the crystals were seen to melt suddenly and then to solidify immediately. The melting of the phase formed by this conversion occurred when the temperature reached 131° C.

Butacaine hydrochloride is *polymorphic*. Phase I, m.pt. 151° C., phase II, m.pt. 131° C., phase III, m.pt. between 110° C. and 120° C. Recrystallisation from different solvents (butanol, pyridine, ethanol-ether, etc.) at room temperature always gave crystals of phase III. Crystals of suitable size for examination in the polarising microscope may be obtained by addition of ether to a solution of the substance in ethanol. The optical crystallographic properties of phase III are reported below.

*Observations in parallel polarised light (crossed Nicols).* Some of the crystals appear as needles and rods which do not extinguish sharply. Extinction dispersion is shown if the crystals are not too thin. Most of the crystals show a lamellar habit with a parallelogram silhouette, often with one or more corners "cut off." They extinguish sharply at an angle of  $24^\circ \pm 1^\circ$ . The elongation is negative.

*Observations in convergent polarised light (crossed Nicols).* On crystals with sharp, oblique extinction a single isogyre is observed in convergent polarised light. The isogyre coincides with the cross hairs when the crystals are in their extinction positions, from which one of the principal indices,  $\alpha$ , and an intermediate refractive index,  $n_1$ , are determined.

*Refractive indices.*  $\alpha = 1.497$ ,  $\gamma = 1.740$  (determined on crushed material). The most frequent value of  $n_1 = 1.703$ .  $\beta$  not determined. The rods without sharp extinction have refractive indices between  $\alpha$  and  $n_1$ .

*Tetracaine Hydrochloride*  
(*Amethocaine Hydrochloride*)

Hydrochloride of 2-dimethylaminoethanol 4-*n*-butylaminobenzoate.

This substance is *polymorphic*. The following melting points are reported in the literature.<sup>13</sup> Phase I, m.pt. 147° C., phase II, m.pt. 139° C., phase III, m.pt. about 130°. Two commercial samples (both pantocain) were used in the melting point determinations.<sup>13</sup> One was a mixture of the phases II and III, the other consisted entirely of phase II. In the present work two samples of B.P. standard were examined ((1) pantocaine, (2) tetracaine hydrochloride). They were both mixtures of the phases II and III.

*Phase II*

Phase II, which is the stable form at room temperature,<sup>13</sup> is obtained by dissolving the substance in ethanol and adding ether (at room temperature) till precipitation occurs. The crystals thus obtained are often too small for examination in convergent light. Well developed crystals may be obtained if the tube containing the solution is placed in crushed ice for about half an hour as soon as the first slight turbidity is observed. If the solution is kept too long at the low temperature, phase III also will crystallise.

*Melting point.* Needles and elongated rods which have been precipitated in this way are seen to melt at 139° C., if they have not been transformed to phase I (m.pt. 147° C.) during the heating. This transformation, accompanied by changes in extinction directions and birefringence, is predominant if the crystals are lying close to each other on the microscope slide.<sup>13</sup>

*Observations in parallel polarised light (crossed Nicols).* Lamellar or needle-shaped rods, always with parallel extinction and positive elongation.

*Observations in convergent polarised light (crossed Nicols).* On lamellar, flattened rods the optic normal interference figure is observed. From this orientation  $\alpha$  and  $\gamma$  are determined. The needles are too narrow to permit observations in convergent light.

*Refractive indices.*  $\alpha = 1.584$ ,  $\gamma = 1.697$ .  $\gamma$  is also observed on the needles (lengthwise); their crosswise index should be near to the  $\beta$  value. The flattened rods may be rolled into the position occupied by the needle-shaped rods and  $\beta$  could thus be estimated, but not exactly measured.  $\beta$  is closer to  $\alpha$  than to  $\gamma$ , optic sign, positive.

*System,* orthorhombic.

*Phase III*

*Preparation.* 1 g. of tetracaine hydrochloride is dissolved in 50 ml. of ethanol (96 per cent.), 50 ml. of ether is added, and the limpid solution is kept at a temperature of 4° to 5° C. for some days (2 to 6 days). Thick,

tabular crystals of parallelogram silhouette grow slowly at the bottom of the solution. Their birefringence is higher than 0.24.

*Melting point.* Crystals prepared in this way, and the corresponding phase in the two commercial samples of tetracaine hydrochloride, are difficult to melt. Between 130° C. and 136° C. they suddenly lose their extremely high birefringence. On further heating the transformed crystals melt at 147° C. (phase I). In crushed material a few small fragments were seen to melt at 134° C. to 135° C. without previous transformation. In mixture with phenazone or acetanilide the highly birefringent crystals of tetracaine hydrochloride gave eutectic points different from the eutectic points of phase II with the same compounds.

*Observations in parallel polarised light (crossed Nicols).* In the commercial samples phase III consists partly of irregular fragments, partly of thick, tabular crystals with parallelogram silhouette. High order white is commonly shown. The relief is high. For the common orientation of the parallelogram tables the extinction is oblique, the angle being about 13° between the vibration direction of the fastest ray and the nearest crystal edge.

*Observation in convergent polarised light (crossed Nicols).* The characteristic common orientation shows a single isogyre, which does not coincide with the cross-hairs for the crystals' extinction positions. No principal refractive indices can be determined from this orientation.

*Refractive indices.*  $\alpha = 1.497$  and  $\gamma > 1.740$ , both determined from crushed material. Most of the crystals showing the tabular habit have their higher index  $> 1.740$ .

In the literature<sup>10</sup> the following refractive indices are reported for tetracaine hydrochloride, without reference to any definite crystallographic phase of this polymorphic substance:  $\alpha = 1.488$ ,  $n_1 = 1.733$ ,  $\gamma > 1.733$ . This high birefringence,  $> 0.24$ , should be found with all commercial samples containing more or less of phase III.

#### *Cocaine Hydrochloride*

*Distinctive optical characters of the sublimate.* When cocaine hydrochloride is heated on the block of a micro melting point apparatus according to the technique described by Kofler,<sup>14</sup> sublimation on the under side of the cover glass starts at temperatures above 160° C. Decomposition sets in at 175° C. to 180° C.<sup>14</sup> The sublimate may be used to advantage for the identification of cocaine hydrochloride by optical crystallographic methods.

*Sublimation procedure.* The substance (1 to 2 mg.) is heated without cover glass until the temperature reaches 160° C. Now a cover glass is placed over the substance with one edge of it supported by a piece of a microscope slide. The temperature is maintained between 165° C. and 170° C. for some minutes. Then the cover glass is removed and placed on another microscope slide for examination, in the polarising microscope, of the crystals attached to it.

*Observations in polarised light (crossed Nicols).* Thin plates without interference colours (grey or white). The following forms are common.

(1) Rectangular or six-sided plates with extinction parallel or symmetric  $\frac{1}{2}$  ( $106^\circ \pm 2^\circ$ ). Elongation negative. In convergent polarised light a centred acute bisectrix is observed. Optic sign, negative. The isogyrs leave the field on rotation of the stage, since  $2V$  is large. From this orientation  $\beta$  and  $\gamma$  is obtained.

(2) Foliate crystals with curved edges. Their orientation is the same as for (1), but their elongation is as a rule positive.

(3) Crystals of variable silhouette, arranged in rosettes. On these crystals a more or less off-centred optic axis figure is observed in convergent light. From the curvature of nearly centred optic axis figures,  $2V$  can be estimated to about  $80^\circ$ . For  $2H$   $87^\circ 57'$  has been reported.<sup>17</sup>

*Refractive indices.* In Keenan's determinative tables<sup>10</sup> the following principal indices are recorded.  $\alpha = 1.570$ ,  $\beta = 1.596$ ,  $\gamma = 1.618$ . From the sublimate  $\beta$  and  $\gamma$  may be determined.  $\gamma$  was found to match the liquid 1.618 and  $\beta$ , the liquid 1.600. The difference between this value (1.600) and 1.596 is without significance, since the observation was made on extremely thin plates.  $\alpha$  could not be observed from any orientation of the sublimate. In the literature different values of the lower index of cocaine hydrochloride<sup>17</sup>:  $\alpha = 1.522$ ,  $\beta = 1.596$ ,  $\gamma = 1.616$  are recorded. From these indices  $2V$  may be graphically computed according to Waldmann's method<sup>15</sup> at about  $55^\circ$ , which could scarcely be the correct angle. In contused material of recrystallised pure cocaine hydrochloride (B.P.), the author was not able to detect refractive indices below 1.570.

Cocaine hydrochloride in some pharmaceutical admixtures may be identified by optical crystallographic examination of the sublimate obtained at  $165^\circ$  to  $170^\circ$  C. by the simple procedure described above. 1 per cent. of cocaine hydrochloride in lactose (about 25 mg.) may be detected.

TABLE I

Substance	Refractive indices		
	$\alpha$	$\beta$ or $n_i$	$\gamma$
1. Butacaine hydrochloride, phase III. . . . .	1.497	1.703 ( $n_i$ )	1.740
2. Tetracaine hydrochloride, phase III . . . . .	1.497	—	> 1.740
3. Tutocaine . . . . .	1.514	—	1.679
4. Larocaine . . . . .	1.521	—	1.742
5. Cinchocaine hydrochloride . . . . .	1.521	—	1.645
6. Procaine hydrochloride* . . . . .	1.542	$\geq 1.560$ ( $\beta$ )	1.742
7. Cocaine hydrochloride . . . . .	1.570	1.596 ( $\beta$ )	1.618
8. Tetracaine hydrochloride, phase II . . . . .	1.584	—	1.697

\* The following principal refractive indices are recorded in U.S.P. XIV:  $\alpha = 1.540$ ,  $\beta = 1.556$ ,  $\gamma > 1.690$ .  $\beta$  was found to be slightly above ( $\geq$ ) 1.560 on fragments showing a centred acute bisectrix (+), and  $\gamma = 1.742$  was determined on contused material.

## SUMMARY

1. The hydrochlorides of 7 commonly used local anæsthetics have been examined in the polarising microscope and their refractive indices are tabulated according to the ascending value of the lower index. The optical crystallographic properties of No. 1, 3, 4, 5 in the table have not been previously reported in literature known to the author. For the

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other substances previously reported data have been completed in this paper.

2. The polymorphism of butacaine hydrochloride and of tetracaine hydrochloride is briefly discussed, and optical data are given for definite crystallographic phases of these drugs.

3. The results are considered useful for micro-identification purposes in pharmaceutical analysis.

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