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Polymorphism of Inclusion Complexes and Unsolvated Hosts. III. Dimorphism of the Alkaloid Colchicine Hydrate. The Structure of Colchicine Monohydrate

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Abstract. The alkaloid colchicine forms, in addition to the previously known dihydrate host–guest complex, a monohydrate complex. The crystal structure of the monohydrate was determined by direct methods and refined to a final R value of 0.046 for 1425 observed reflections. Crystal data are: orthorhombic, space group $P2_12_12_1$, a = 9.145(2) Å; b = 13.270(3) Å; c = 17.942(4) Å, V = 2177(1) Å^3, Z = 4, D_x = 1.22 g cm^{-3}, T = 293 K. The conformation of the molecule is practically identical with the conformation in the dihydrate inclusion complex. Water molecules show proton donor as well as proton acceptor properties and are hydrogen bonded with the three colchicine molecules giving rise to the three dimensional H-bonded network.

Key words: Crystal structure, X-ray crystallography, colchicine, monohydrate, H-bond.

Supplementary data relevant to this article have been deposited with the British Library as Supplementary Publication No. SUP 82215 (7 pages).

1. Introduction

Colchicine is an alkaloid isolated from plants of the *Colchicum* and *Merendera* species. It shows different types of biological action, the main ones being its arrest of mitosis [1] as well as its property of inducing polyploidy in plants [2].

The first X-ray diffraction study of colchicine was performed by King *et al.* [3] on the colchicine–methylene bromide complex. This work provided confirmation of the molecular structure and a very rough geometry of colchicine since the limited amount of X-ray data did not give a complete three-dimensional structure. This was first established by the X-ray study of Lessinger and Margulis [4] carried out on crystals of colchicine dihydrate which are easily grown by evaparation from an aqueous solution containing tris(hydroxymethyl)-aminomethane (tris buffer). However, as shown by our experiments, the dihydrate of colchicine is an unusual form, since under ordinary conditions, colchicine crystallizes in a monohydrate

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modification from aqueous solutions. As the crystal structure of this conventional form of colchicine has not been reported we carried out an X-ray structure analysis of colchicine monohydrate and report a comparative discussion of the two structures.

2. Experimental

Single crystals of colchicine monohydrate were obtained from a solution of colchicine in aqueous ethanol under ambient conditions. A crystal of approximate dimensions 0.2 × 0.3 × 0.6 mm was used for measurements on a Syntex-$P2_1$ diffractometer. The lattice parameters were determined by a least-squares fitting of the setting angles of 15 reflections (2θ in the range 20–30°). The crystal data are: $C_{22}H_{25}NO_6 \cdot H_2O$, orthorhombic, space group $P2_12_12_1$, a = 9.145(2) Å; b = 13.270(3) Å; c = 17.942(4) Å, V = 2177(1) Å^3, Z = 4, D_x = 1.22 g cm^{-3}, T = 293 K.

The intensities of reflections were measured with graphite monochromated CuK_α radiation ($\lambda = 1.54$ Å) to $2\theta_{\max} = 120°$ using the $\theta/2\theta$ scan technique. No significant intensity variation was observed for two standard reflections monitored after each group of 100 reflections. Data were corrected for Lorentz and polarization effects but not for absorption. Out of 1760 measured reflections 1425 had $I > 2\sigma(I)$ and were used for further calculations. The structure was solved by direct methods with the program SHELXS-86 [5]. Colchicine and water molecules were located on the 'best' E map. The structure was refined first with isotropic and then with anisotropic thermal parameters by least-squares methods using the program SHELX-93 [6]. Hydrogen atom positions were determined from the ΔF maps and their positional and isotropic thermal parameters were included in the refinement. Final values of R and WR are 0.046 and 0.047, respectively. In the final ΔF map, calculated after the last cycle of refinement, the maximum and minimum peaks were 0.60 and $-0.52e$ Å^{-3}, respectively.

3. Results and Discussion

The atom parameters are given in Table I. Tables of H atom coordinates, anisotropic thermal parameters and structure factors are in the Supplementary Material. The numbering scheme of colchicine molecules is shown in Figure 1.

3.1. Molecular structure

The colchicine molecule comprises three condensed rings with different substituents (Figure 1), i.e. a six-membered aromatic ring A with three methoxy-groups, a cycloheptadiene ring B with an acetylated amino-group and a seven-membered ring C with carbonyl and methoxy-groups (troponoid ring).

Atoms of the A ring are coplanar within 0.02 Å and the methoxy group O(5)—C(20) is situated almost parallel to the ring while the two other methoxy groups

Table I. Atom coordinates ($\times 10^4$) and temperature factors (Å^2 $\times$ 10^3).

Atom	x	y	z	U^*
C(1)	1206(10)	3638(6)	14020(5)	57(3)
C(2)	2214(9)	4324(6)	13627(4)	51(3)
C(3)	2812(9)	4211(6)	12935(4)	51(3)
C(4)	2635(8)	3414(5)	12408(8)	46(3)
C(5)	1736(8)	2578(5)	12409(4)	42(2)
C(6)	658(8)	2360(5)	12973(4)	43(2)
C(7)	469(9)	2830(6)	13650(4)	47(2)
C(8)	1909(8)	1834(5)	11781(4)	43(2)
C(9)	2141(8)	811(5)	11941(4)	48(2)
C(10)	2355(10)	127(6)	11363(4)	54(3)
C(11)	2319(10)	442(5)	10620(4)	56(3)
C(12)	1957(8)	1434(6)	10460(4)	46(3)
C(13)	1772(8)	2129(5)	11037(4)	45(2)
C(14)	2057(9)	438(6)	12738(4)	50(3)
C(15)	447(9)	472(5)	13014(4)	48(3)
C(16)	−334(8)	1455(5)	12802(4)	43(2)
C(17)	−2807(9)	2151(6)	12961(4)	54(3)
C(18)	−129(9)	3246(6)	10789(5)	60(3)
C(19)	707(12)	1340(7)	9274(5)	79(4)
C(20)	2957(15)	−1188(6)	10135(6)	97(5)
C(21)	3506(11)	5905(7)	13849(5)	76(4)
C(22)	−4271(9)	1999(8)	13347(6)	75(4)
N(1)	−1756(7)	1481(5)	13170(3)	45(2)
O(1)	968(8)	3776(5)	14696(3)	86(3)
O(2)	2551(8)	5117(4)	14072(3)	70(2)
O(3)	1437(6)	3111(3)	10855(3)	48(2)
O(4)	1847(6)	1789(4)	9731(3)	56(2)
O(5)	2602(8)	−156(4)	10012(3)	73(2)
O(6)	−2574(7)	2810(4)	12480(3)	67(2)
O(W)	2349(8)	4939(4)	15780(3)	83(2)

* $U = \frac{1}{3}\Sigma\Sigma U_{ij} a_i^* a_j^* \mathbf{a}_i \cdot \mathbf{a}_j$.

are located nearly perpendicular to it. The only asymmetric atom of the molecule C(16) is in ring B to which the N-acetyl group is attached in an equatorial position. The N-acetyl side chain consisting of atoms C(22), C(17), O(6) and N(1) is nearly planar within 0.05 Å.

The troponoid ring C is roughly planar within 0.09 Å. Long and short bonds clearly alternate in this ring. The methoxy group O(2)—C(21) is located nearly parallel to the ring. The dihedral angle between rings A and C is equal to 54.3(2)° and these rings are twisted about the C(5)—C(8) bond. All of these show that

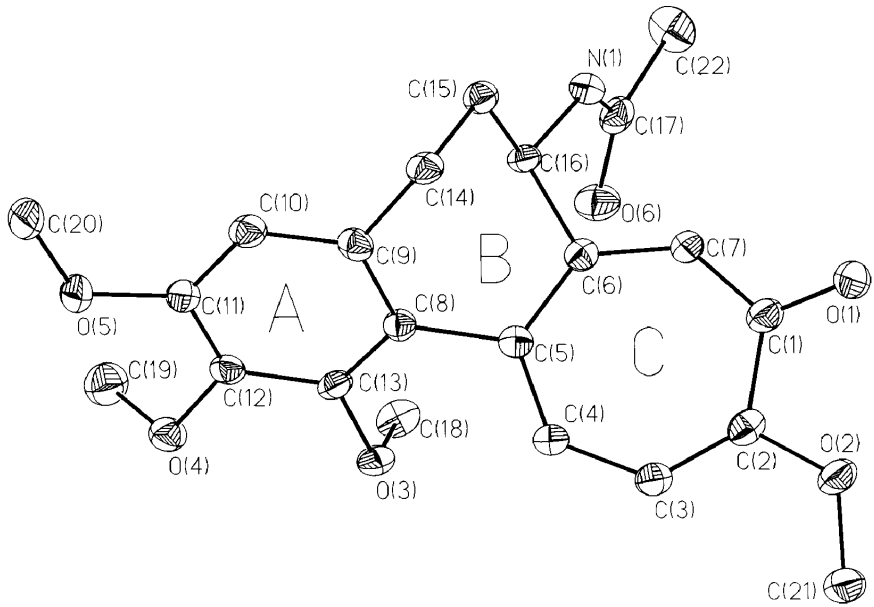

Figure 1. The colchicine molecule and its numbering scheme.

the conformation of the colchicine molecule in the monohydrate is practically identical to the conformation in the dihydrate complex and in the other derivatives of colchicine [7, 8].

3.2. CRYSTAL STRUCTURE

In the structure of colchicine dihydrate alkaloid molecules are associated via N(1)—H···O(1) H-bonds into columns around the 2_1 screw-axis. Wide, tubular empty regions, which should be filled by water molecules, are formed during the packing of such columns. Each of the four water molecules located in this area are H-bonded to others, as well as to host molecules. The peculiarity of the colchicine dihydrate is the existence of separate water regions in the structure [4]. It should be noted that water regions also exist in the structure of racemic colchicine dihydrate [9]. The structure of the (±)-colchicine dihydrate can be described as that of chains of alternate D and L molecules related by a glide plane and hydrogen bonded by the N—H···O(1) interaction.

In the case of colchicine monohydrate the water molecule is H-bonded to the three different alkaloid molecules showing proton donor as well as proton acceptor properties (Figure 2). The geometry of H-bonds O(W)—H···O(1), O(W)—H···O(3)^{i} and N(1)—H···O(W)^{ii} is given in Table II. Each colchicine molecule is H–bonded with three water molecules, giving rise to a three-dimensional H-bonded network. There is no direct interaction between colchicine molecules in the struc-

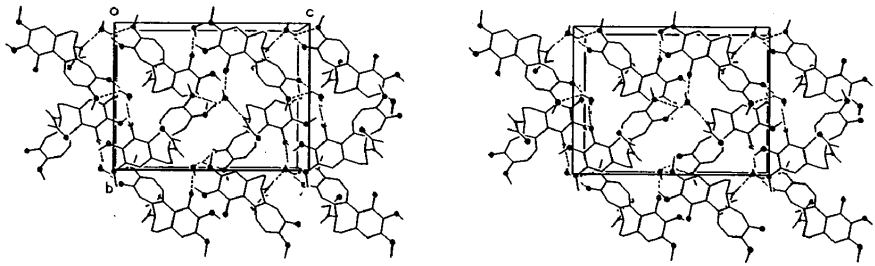

Figure 2. Crystal structure of colchicine monohydrate.

Table II. Intermolecular hydrogen bonds in the colchicine monohydrate structure.

Bond	D···A (Å)	D—H (Å)	H···A (Å)	∠D—H···A (°)
O(W)—H···O(1)	2.79	0.98	1.88	155
O(W)—H···O(3)^{i}	2.82	0.82	2.06	154
N(1)—H···O(W)^{ii}	2.79	1.01	1.79	168

Symmetry codes: (i) $0.5 - x$, $1 - y$, $0.5 + z$; (ii) $0.5 + x$, $0.5 - y$, $3 - z$.

ture and the same is true for the water molecules. In the strict sense this complex of colchicine with water is distinct from the dihydrate, i.e. it is not a host–guest type complex.

Thus colchicine gives two molecular complexes with water, depending on the crystallization conditions. In the structure of the dihydrate separate water regions exist while in the crystals of the monohydrate alkaloid and solvent molecules are evenly distributed. If water molecules are absent from the crystallization media an amorphous powder of colchicine is formed.

References

1. D. Soifer (Ed.): *The Biology of Cytoplasmic Microtubules*, Ann. N.Y. Acad. Sci. 253, esp. pp. 213–231 (1975).
2. O. J. Eigsti and P. Jr. Dustin: *Colchicine in Agriculture, Medicine, Biology and Chemistry*, Iowa: Iowa State College Press (1955).
3. M. V. King, J. L. deVries and R. Pepinsky: *Acta Crystallogr.* **5**, 437 (1952).
4. L. Lessinger and T. N. Margulis: *Acta Crystallogr. B* **34**, 578 (1978).
5. G.M. Sheldrick: *Acta Crystallogr. A* **46**, 467 (1990).
6. G.M. Sheldrick: SHELXL-93: *Program for the Refinement of Crystal Structures*, University of Göttingen, Germany (1993).
7. L. Lessinger and T. N. Margulis: *Acta Crystallogr. B* **34**, 1556 (1978).
8. T. N. Margulis: *J. Am. Chem. Soc.* **96**, 899 (1974).
9. R. Dumont, A. Brossi and J. V. Silverton: *J. Org. Chem.* **51**, 2515 (1986).