CRYSTALLINE CONFORMATION OF ENNIATIN B

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1. Introduction

The spatial structure of the depsipeptide ionophore-enniatin B (fig.1) has been studied theoretically by semiempirical [1] and quantum-chemical PCILO-methods [2] as well as by solution techniques [3–8]. Two principal conformations were found: a C_3-symmetric form called *P* and a non-symmetric form called *N*, predominant in polar and non-polar solvents, respectively. The *P*-form is also realized in complexes with alkali and alkaline earth metal ions in solutions [3,6] and in crystals [9]. This communication presents results of the X-ray structural analysis of enniatin B crystals obtained from acetonitrile in an attempt to isolate the (Enniatin B)$_2$·KNCS 'sandwich' complex [10].

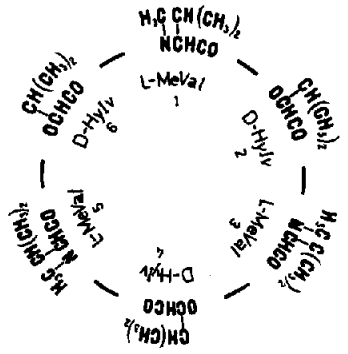

Fig.1. Formula of enniatin B.

2. Materials and methods

The crystals are triclinic prisms, space group P_1, $a = 9.83$ Å, $b = 9.69$ Å, $c = 9.83$ Å, $\alpha = 92°25'$, $\beta = 99°27'$, $\gamma = 92°25'$, with $Z = 1$, $D_x = 1.15$ gm·cm^{-3}. The intensity data were collected by means of an Inraf-Nonius semi-automatic difractometer using CuK_α and the structure resolved by the Sayre-Zachariasen-Cochan statistical method [11–13] using the Röntgen-70 programs [14] developed for the M-220 M computer. The structure determination was made assuming a P_1-space group based on the fact that the statistical values $|E^2| = 1.020$, $|E^2-1| = 0.948$ and $|E| = 0.828$ were close to the theoretical ones $|E^2| = 1.000$, $|E^2-1| = 0.968$ and $|E| = 0.798$ for the centrosymmetrical case and that the molecule has a pseudocenter of symmetry. The R-factor as calculated from the coordinates of the 20 strongest peaks in the E-synthesis had a value of 0.42. In the first Fourier synthesis made on the basis of the P_1-space group, i.e. a symmetrized molecule with statistical distribution of the N-CH_3 and O′groups besides localization of all the non-hydrogen atoms three extra peaks attributed to non-existent methyl carbons were revealed. In the course of the structural study preference was given to one of the two possible arrangements of the O′ and N atoms in the main chain, because this led to a lower R-factor and better interatomic distances. The final model of the structure was arrived at by successive Fourier syntheses in the non-centrosymmetric approximation.

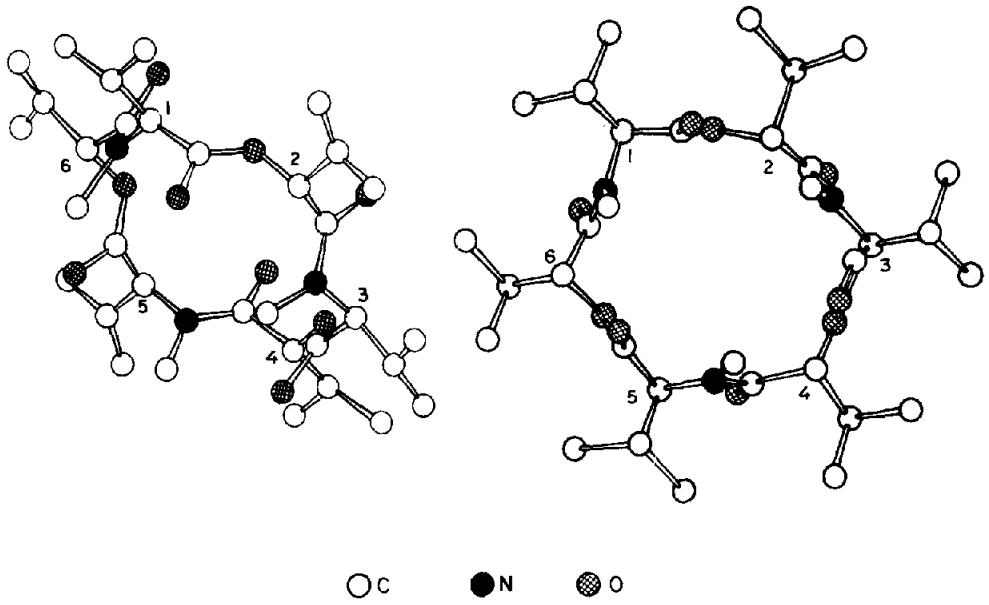

Fig.2. The crystalline conformation of enniatin B. (a) Projection on the XZ-plane. (b) View down the pseudo-three-fold axis.

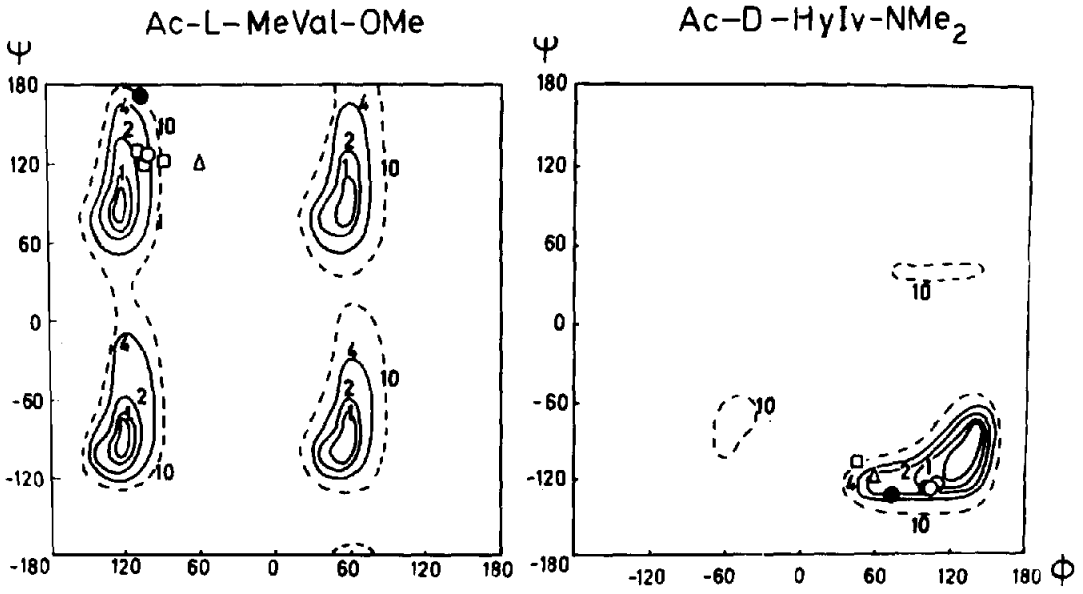

Fig.3. Comparison of parameters for the crystalline and *P* conformations of enniatin B as points on the conformational maps for the amino and hydroxy acid fragments. (○) X-ray data; (△) X-ray data for the K^+ complex [9]; (●) semiempirical calculation for the *P* conformation [1]; (□) quantum-chemical calculation for the *P* conformation [2].

The least-squares refinement of the atomic positions and isotropic thermal parameters was made until ultimate reduction of the R-factor to 0.12 for 1700 reflections.

3. Results and discussion

The structure obtained is shown in fig.2, its conformational parameters as calculated from the atomic coordinates are listed in the table.

The ϕ and ψ angles fall into the allowed regions of the Ac-L-MeVal-OMe and Ac-D-HyIv-NMe_2 conformational maps [15], compounds modelling the amino and hydroxy acid residues of antibiotic. For comparison, values obtained by theoretical analysis of the antibiotic and by incomplete X-ray analysis of the K^+ complex [9] are also given (fig.3).

The structural parameters of the molecule with its center of pseudosymmetry and pseudo-C_3-axis are quite close to the earlier discussed form *P* (see table 1) with its characteristic up-down alteration of the carbonyls with respect to the average plane of the ring. The N-methylamide and ester groups possess the trans configuration but according to the ω, θ_N and θ_C, parameters [16,17] deviate noticeably from planarity.

The structure has a molecular cavity of ~2.7 Å diameter as determined from the distance of the pseudocenter to the N_1, N_2 and N_3 atoms (3.04, 3.17 and 2.94 Å respectively) and a van der Waals radius of the N atoms (1.58 Å). The corresponding distances of the main chain O′ atoms are 3.00, 3.25 and 3.27 Å, and of the carbonyl O atoms are 3.58, 3.61, 3.68, 3.72, 3.54 and 3.84 Å. The pseudo-equatorial isopropyl side chains lend lipophilicity to the molecular periphery. The optimal steric requirements of the molecular substituents cause a slight relative and opposite turning of neighbouring isopropyl groups along the C^α–C^β bound with respect to the average molecular plane. The $C^\alpha H$–$C^\beta H$ protons are in the energetically advantageous trans position in both the amino and hydroxy acid residues.

The study provides the first accurate determination of an enniatin ionophore. The aforementioned X-ray analysis of the (enniatin B)·KI complex [9] was not carried out to determination of the tridimensional coordinates of the atoms. Hamilton et al. [18] recently reporting the three dimensional structure of the complex $(beauvericin)_2$·$(barium\ picrate)_2$ also do not give values for the atomic coordinates or torsion angles.

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Table 1
Standard conformational parameters of enniatin B (in degrees)

Residue number	ϕ	ψ	ω	θ_N	θ_C'	$\chi^{1,1}$	$\chi^{1,2}$
1	−104.00	123.23	−172.16	12.25	23.60	−65.00	180.00
2	110.75	−125.90	−180.00		14.00	167.34	55.00
3	−100.30	128.60	−175.00	15.33	14.66	−44.66	180.00
4	104.60	−126.35	175.66		12.00	170.50	52.66
5	−108.75	131.50	169.16	16.83	17.16	−50.66	173.84
6	106.10	−126.50	171.50		12.50	−176.34	74.66

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