CONSTITUTION AND ABSOLUTE STEREOCHEMISTRY OF THE ANTIBIOTIC SARUBICIN A

K. ECKARDT,[a†] D. TRESSELT,[a] W. IHN,[a] M. KAJTÁR,[b†] J. ÁNGYÁN,[c] L. RADICS[d] and M. HOLLÓSI[b]

[a]Akademie der Wissenschaften der DDR, Forschungszentrum für Molekularbiologie und Medizin, Zentralinstitut für Mikrobiologie und experimentelle Therapie, DDR-6900 Jena, Beutenbergstr. 11, DDR
[b]Institute of Organic Chemistry, Eötvös University, P.O. Box 325, H-1445 Budapest, Hungary
[c]CHINOIN Pharmaceutical and Chemical Works Ltd., P.O. Box 110, H-1325 Budapest, Hungary
[d]Central Research Institute for Chemistry, P.O. Box 17, H-1525 Budapest, Hungary

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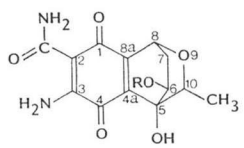
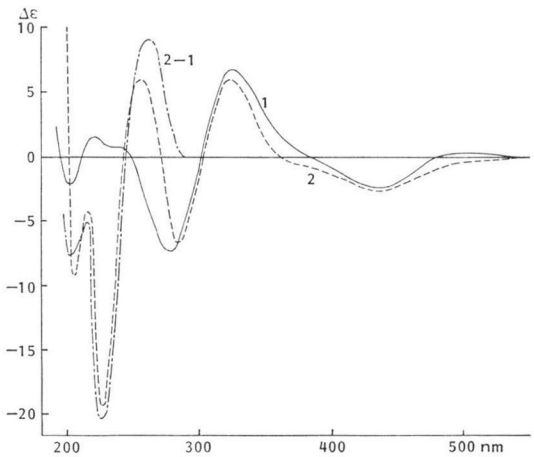
By spectral (UV-VIS, IR, NMR, MS and CD) methods the quinone antibiotics sarubicin A and U-58,431 were shown to have identical constitution and stereochemistry. Chiroptical data and their theoretical analysis have settled the common absolute configuration as 5*S*, 6*R*, 8*R*, 10*R*.

Recently we reported on the isolation of sarubicin A, a novel quinone antibiotic and described its constitution as inferred from NMR and mass spectral data[1,2]. In the proposed structure, however, the position of the $CONH_2$ and NH_2 groups (both attached to the quinone moiety), as well as the stereochemistry of the entire molecule remained to be established. Shortly after the appearance of our paper, SLECHTA and coworkers[3] have published the structure and X-ray-based relative configuration of another quinone antibiotic, U-58,431. Inspection of the structural features and available physico-chemical properties suggested the existence of a close relationship between the two compounds. In fact, subsequent direct comparison of the UV-VIS, IR, NMR and mass spectra of sarubicin A and antibiotic U-58,431* disclosed their constitutional identity; consequently, sarubicin A is represented by formula **1**.**



1 R = H
2 R = COC_6H_5

Fig. 1. CD spectra (in ethanol) of sarubicin A and antibiotic U-58,431 (**1**), of 6-*O*-benzoylsarubicin A (**2**) and the difference spectrum (**2**−**1**).



† To whom communications should be addressed.

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** In formula **1**, we use our earlier[1,2] numbering system which is different from that used by SLECHTA *et al.*[3]

The CD spectra recorded in ethanol of sarubicin A and antibiotic U-58,431 have been found to be perfectly identical (Fig. 1) showing that the two substances also possess a common absolute stereochemistry.

In this paper we report on the results of our chiroptical studies devoted to determining the absolute stereochemistry of sarubicin A.

Contrary to cases of more familiar chromophores, no empirical rules are known as yet that correlate the sign pattern of CD bands due to the quinone chromophore with the geometry of its chiral environment. Therefore the absolute stereochemistry of sarubicin A cannot be deduced directly from its CD spectrum. With the relative configurations already established[3], however, the exciton chirality method of HARADA and NAKANISHI[4,5] promised to offer a simple solution to the problem. The application of this technique required the conversion of the parent antibiotic into its benzoyl derivative.

6-*O*-Benzoylsarubicin A (**2**)

Sarubicin A (3 mg) dissolved in pyridine was treated with benzoyl chloride at room temperature. The evaporated reaction mixture was chromatographed on a silica-gel plate (solvent mixture: benzene - acetic acid, 7: 3), and the major product (Rf 0.7) isolated. Its UV spectrum was almost identical with that of the parent compound with an additional band at 230 nm. From the intensity of the latter (ε 13,000, in addition to the original spectrum), the presence of one benzoyl group in the molecule could be inferred. The location of the benzoyl group at the O(6) atom followed from the ^{1}H NMR spectrum displaying a 1.2 ppm downfield shift for the resonance of the C(6)-H proton with respect to its value in the ^{1}H NMR spectrum of sarubicin A[2].

The CD spectrum of the 6-*O*-benzoyl derivative **2** is very similar to that of sarubicin A above 300 nm; whereas it displays a characteristic exciton couplet superimposed on the CD spectrum of the parent compound between 300 and 200 nm (Fig. 1). The resulting difference spectrum (also shown in Fig. 1) exhibits a positive maximum at 260 nm and two negative bands at 225 and 208 nm.

Calculations

The geometry of sarubicin A is sufficiently rigid to remain practically unchanged upon introduction of the benzoyl group in the molecule. It is therefore reasonable to assume that the optical activity resulting from the exciton interaction between transitions of the benzoate and the quinone chromophores is superimposed on the optical activity of the parent molecule in a simple additive manner. Thus the calculations can be confined to the interpretation of the difference spectrum within the approximation of the exciton chirality approach[4,5].

The transitions of the quinone chromophore have been calculated for the achiral 3-amino-2-carbamoyl-1,4-benzoquinone moiety of sarubicin A using CNDO/S-CI approximation[6] and the X-ray-based geometry of reference 3. Despite some inconsistencies observable in the sequence of energy levels, the calculated transitions in Table 1 are in reasonable agreement with experimental UV and CD data. The charge transfer transition[4] of the benzoate chromophore found at 230 nm in the UV spectrum of **2** is expected to exsert the strongest coupling with the two intense and closest to it in energy $\pi\rightarrow\pi^*$ transitions of the quinone chromophore (which appear as a single band at 212 nm in the UV spectrum). Based on this assumption, the positive maximum at 260 nm in the difference CD spectrum has been assigned to the benzoate transition, whereas the two negative bands at 225 and 208 nm have been attributed to the calculated highest energy $\pi\rightarrow\pi^*$ transitions of the quinone chromophore.

Table 1. Calculated spectroscopic data of 3-amino-2-carbamoyl-1,4-benzoquinone and experimental UV-VIS and CD spectra of sarubicin A (**1**) and its 6-*O*-benzoyl derivative (**2**).

Calculation			Experiment			
Type of transition	f	λ (nm)	UV-VIS (EtOH) λ (nm) ($\varepsilon \times 10^{-3}$)		CD (EtOH) λ (nm) ($\Delta\varepsilon$)	
			1	**2**	**1**	**2**
$n\rightarrow\pi^*$	0	450	469 (1.67)	478 (1.60)	502 (+0.28)	437 (−2.64)
$n\rightarrow\pi^*$	0	416			434 (−2.25)	
$n\rightarrow\pi^*$	0	326	294 (6.07)[a]	294 (5.80)[a]	324 (+6.68)	321 (+6.05)
$n\rightarrow\pi^*$	0	240[b]	272 (11.87)[a]	272 (12.40)[a]	278 (−7.46)	284 (−6.80)
$\pi\rightarrow\pi^*$	0.12	321[b]	263 (13.54)	263 (14.35)		
				230 (18.60)		255 (+6.00)
$\pi\rightarrow\pi^*$	0.14	215	212 (17.68)	214 (22.40)	236 (+0.67)[a]	227 (−19.40)
					220 (+1.45)	
$\pi\rightarrow\pi^*$	0.66	205			201 (−2.12)	207 (−9.70)

[a] Shoulder.

[b] The sequence of the two transitions is reversed.

Table 2. Calculated exciton optical activity of 6-*O*-benzoylsarubicin A

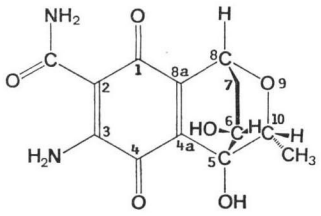
Chromophore	λ (nm)	Rotational strength in 10^{-40} c.g.s. units calculated for torsion angle of			
		0°	+60°	+90°	180°
Benzoate	230	+12.1	+8.8	+7.0	+2.1
Quinone	215	−3.1	−1.6	−0.8	+0.8
Quinone	205	−9.0	−7.2	−6.2	−2.9

To calculate the exciton optical activity of the benzoyl derivative **2**, a benzoate group of standard geometry has been generated in lieu of the OH group at position 6 of the parent molecule, and the calculations have been performed for four rotational conformations of the benzoyl group with the C(5)C(6)–O(6)C(=O) torsion angle assuming 0°, +60°, +90° and 180°, respectively. The direction of the electric transition moment due to the intramolecular charge transfer transition was taken along the long axis of the benzoate chromophore[4], whereas calculated directions were used for the $\pi\rightarrow\pi^*$ transitions of the quinone chromophore. The point dipoles of the electric transition moments were placed in the geometric centres of the benzene and quinone rings, respectively.

Summarized in Table 2 are the calculated rotational strengths obtained by considering the dipole-dipole interactions between the three electric transition moments mentioned above and by assuming *R* configuration for C(6) (as portrayed for the enantiomer of U-58,431 in Fig. 3 of reference 3). In view of the approximations used in describing the chromophores and the limited number of transitions included in the calculations, only the signs of the calculated rotational strengths can be considered as significant. Inspection of the data in Table 2 reveals that, for three of the four rotational conformations of the benzoyl group considered, the sign patterns of the calculated exciton bands agree with the experimental sign pattern displayed in the CD difference spectrum (see Fig. 1). The major differences between experimental and calculated intensities of the exciton bands are presumably due to the neglect of the coupling between the quinone $\pi\rightarrow\pi^*$ and the higher energy B-type transitions of the benzoate chromophore in these calculations.

The agreement between the experimental and calculated sign patterns indicates that carbon atom C(6) in sarubicin A (and, consequently, in antibiotic U-58,431) has *R* absolute configuration which,

Fig. 2. Absolute configuration of sarubicin A (U-58,431).



on the basis of the X-ray-derived relative configurations[3] of the other chiral centres of the molecule, settles the absolute geometry of the antibiotic as 5*S*, 6*R*, 8*R*, 10*R* (Fig. 2).

It is interesting to note that the absolute configuration thus obtained is identical with that of the corresponding moiety of granaticin, a related quinone antibiotic[7].

After completion of this work we learned about the results of comparative chiroptical studies of B. KRONE who also found that the common chiral fragments of granaticin and sarubicin A are of the same absolute configuration. (Personal communication of Dr. B. KRONE, University of Göttingen, F.R.G.)

Experimental

^{1}H NMR spectra were obtained at 100 MHz in $CDCl_3$ solution using Varian XL-100/15 FT NMR spectrometer. UV-VIS and CD spectra were measured on Specord M 40 (Carl Zeiss, Jena) and Roussel-Jouan Dichrograph No III (Jobin-Yvon) instruments, respectively.

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