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Circular Dichroism of Michellamines: Independent Assignment of Axial Chirality by Calculated and Experimental CD Spectra[1]

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Abstract: The circular dichroic behavior of the michellamines, dimeric naphthylisoquinoline alkaloids, was investigated. Due to the molecular size and conformational flexibility, which makes direct calculations of the CD spectra very difficult, and because of the lack of direct standards for an empirical comparison, there is no simple possibility for the interpretation of the CD spectra of such quateraryls. The chiroptical contributions of the particular stereogenic elements were shown to behave largely additively and thus allow comparison of spectra of the quateraryls with those of the corresponding 'monomeric' naphthylisoquinolines. This comparison was performed first of all with the experimental and theoretical CD spectra of ancistrobrevine B, then with the predicted spectra of the authentic molecular 'halves' of the michellamines.

INTRODUCTION

Ancistrocladus korupensis, a Central African liana endemic to Cameroon, is a 'new' Ancistrocladaceae species recently detected in an anti-HIV screening program initiated by the US National Cancer Institute. From this plant, remarkable quateraryl alkaloids, the michellamines, were isolated.[2] These novel natural products were found to have the joint constitution **1**, highly polar with their six free phenolic groups and the two amino functions. They are unprecedented constitutionally symmetric dimers of a 5,8'-coupled naphthylisoquinoline; the formal monomer is very similar to natural ancistrobrevine B (**2**), which was isolated, in parallel investigations, from *A. abbreviatus*.[3] Apart from the unusual constitution of the michellamines, the stereostructures are also challenging: there are four stereogenic centers, two stereogenic biaryl axes, and a configuratively labile central biaryl axis between the two molecular parts. The relative configuration at centers and axes was elucidated by NMR spectroscopy.[2] Likewise from

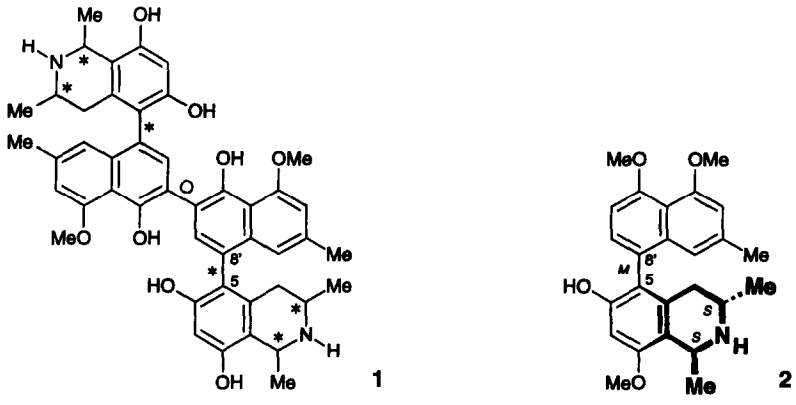


Fig. 1. Constitution **1** of the michellamines A – C, structure of the related naphthylisoquinoline alkaloid ancistrobrevine B (**2**); stable stereogenic elements (centers or axes) are denoted by '*' labile ones by 'o'.

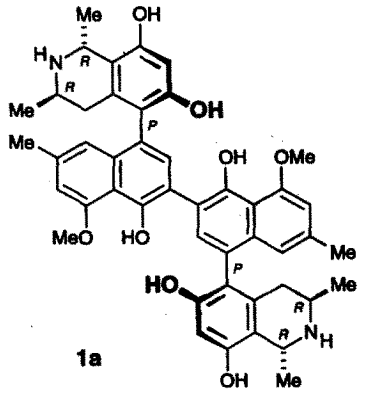
NMR studies it was manifest that michellamines A and C consisted of two spectroscopically identical (*i.e.* homomorphous or enantiomorphous) halves, whereas michellamine B could clearly be shown to be built up from two spectroscopically different (*i.e.* diastereomorphous) molecular parts.

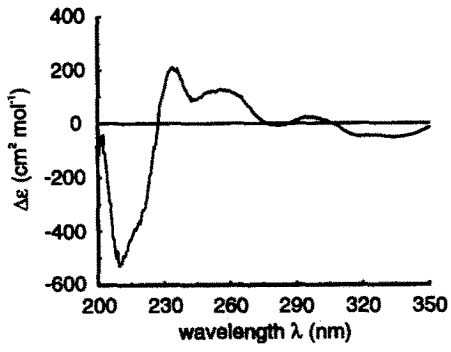
The absolute configuration at the stereocenters was determined by a ruthenium-mediated oxidative degradation reaction, leading to enantiomerically pure *R*-3-aminobutyric acid and D-alanine, thus establishing all the stereocenters to be *R*-configurated.[4,5] From this information, with the known relative configurations at centers *vs.* axes as deduced from NMR, the absolute configurations at the axes had also been concluded.[4,5] Accordingly, the 3 michellamines A, B, and C are represented by the stereostructures **1a**, **1b**, and **1c** (Fig. 2). Hence, all these compounds are stereochemically identical at the stereocenters and represent a complete series of atropodiastereomers. Because of the importance of an absolutely reliable knowledge of the complete stereostructure of these novel quateraryls, we now report on an independent confirmation of the axial configuration by comparison of experimental and computational CD spectroscopy.

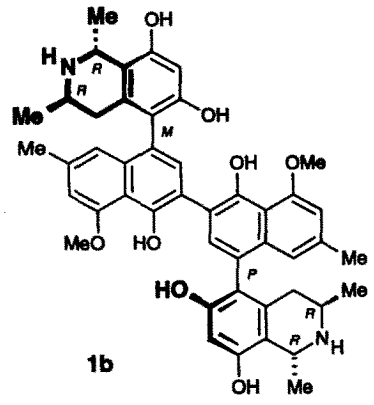
RESULTS AND DISCUSSION

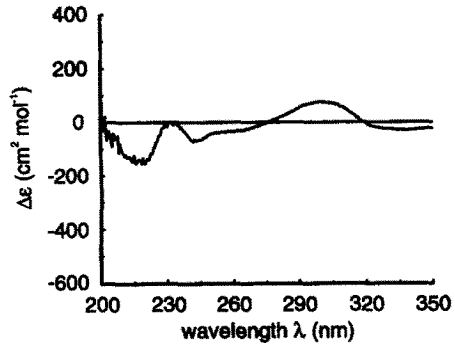
1. CD Spectra of the Michellamines:

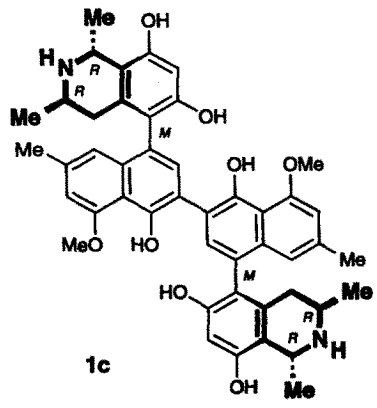
For such an independent proof, the CD option seemed ideal. Fig. 2 shows the CD spectra of michellamines A, B, and C. As expected, one finds nearly opposite CD curves for the michellamines A (**1a**) and C (**1c**), completely in agreement with their opposite axial configurations. By contrast, the curve for michellamine B (**1b**), with its two heterochiral axes, is, as expected, far less intensive, but by no means flat, since the molecule is still chiral and remains under the influence of the stereogenic centers, which are all *R*-configurated. Yet, besides these more *qualitative* rationalizations, an unambiguous independent proof of the concluded absolute configurations is very difficult both empirically, because of the completely unprecedented structural type, *and* computationally, because of the now drastically larger molecular size. For this reason, it was important to know whether, at all, the two molecular moieties influence each other











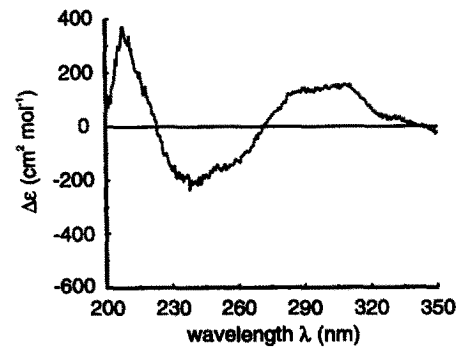


Fig. 2. Stereostructures and CD spectra of michellamines A – C; for UV data, see Experimental.

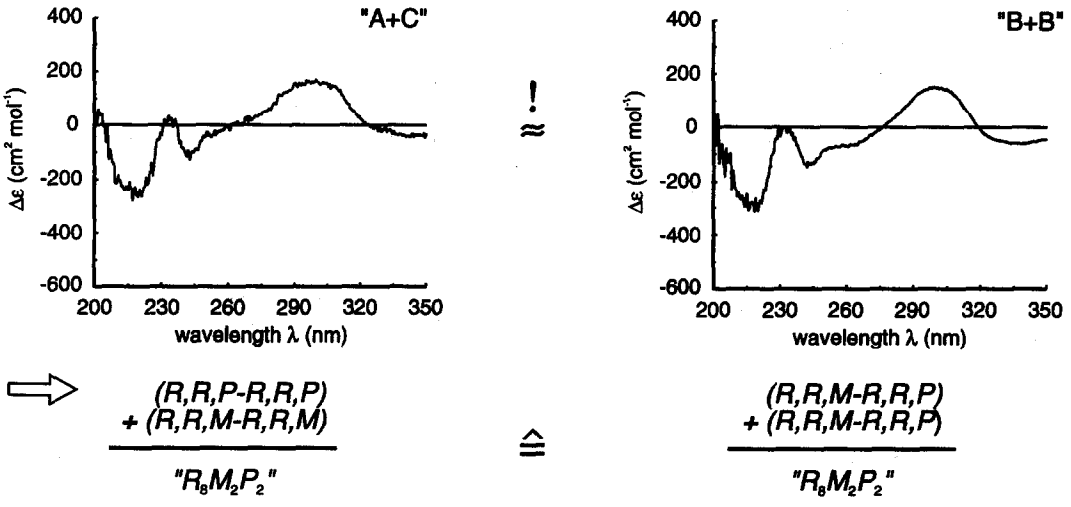


Fig. 3. Numerically added CD spectra composed of michellamines A (**1a**) + michellamine C (**1c**) (left) and 'doubled' spectrum of michellamine B (**1b**) (right).

chiroptically or whether one might (hopefully) just consider the molecular halves and perform empirical comparisons or theoretical calculations with such monomers. A first answer to the question whether the chiroptical contributions of the different stereoelements would behave additively or not might be seen for instance if, purely numerically, the addition of the spectra of michellamines A (**1a**) plus C (**1c**), formally consisting of the stereo-contributions '*R,R,P-R,R,P*' (from **1a**) + '*R,R,M-R,R,M*' (from **1c**) would result in the same overall spectrum as if the spectrum of michellamine B (**1b**) would just be doubled, because in each case, the same overall number of correspondingly configurated stereocenters and axes (namely '$R_8M_2P_2$') would additively contribute to the overall spectrum. This additivity of the stereocontributions is really the case to an astonishingly significant degree (see Fig. 3), so that one may expect that possibly the 'halves' of the michellamines also give independent chiroptical contributions.

2. Comparison with the Experimental and Calculated CD Properties of Ancistrobrevine B (2):

Hence, in a first approach it seemed appropriate to compare the hypothetical michellamine 'halves' empirically with the experimental spectra of known 'monomeric' alkaloids or, theoretically, with the predicted spectra of such halves. Such a very closely related 'monomeric' naphthylisoquinoline alkaloid is the aforementioned ancistrobrevine B (**2**).[3] With its 1*S*,3*S*-configuration, and the axis *M*-configurated, it would be, in an *O*-demethylated form, exactly the enantiomer of the molecular 'half' of michellamine A. And indeed, the CD curve (Fig. 4) is, to a high degree, near-opposite to that of michellamine A (**1a**) in the 200-260 nm region, whereas, as expected, it resembles, in part, that of michellamine C (**1c**), which has identical axial (though opposite central) configurations. This structural attribution is further

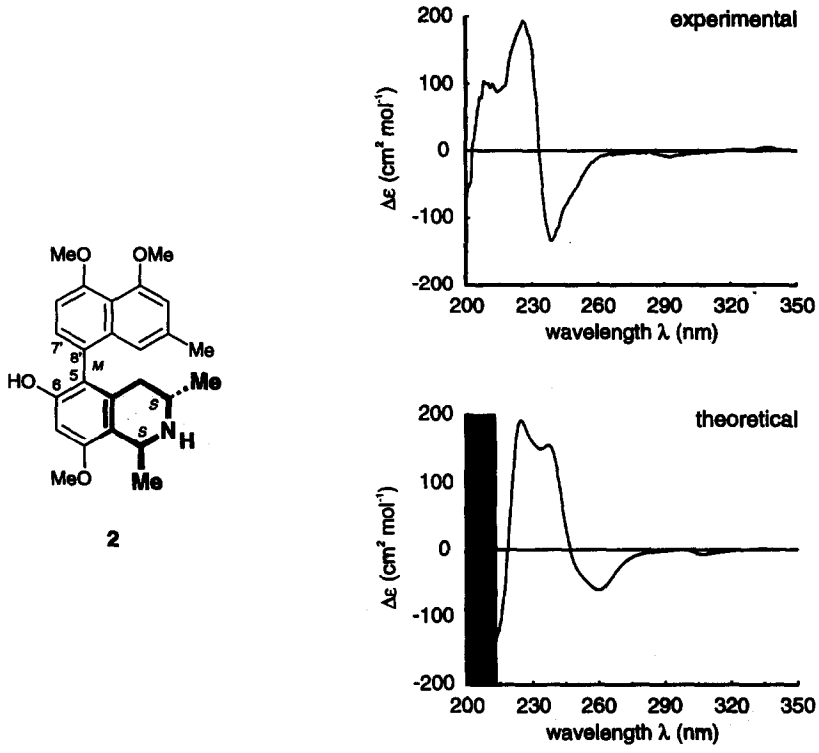


Fig. 4. Experimental and theoretical CD spectra of natural ancistrobrevine B (**2**); the systematically occurring shift difference of experimental and theoretical spectra[6] of ca. 14 nm is 'optically subtracted' by hatching.

underlined by the result of the theoretical calculation of the CD spectrum of ancistrobrevine B (**2**) using a CNDO/S → Boltzmann-weighting approach introduced into the computational prediction of CD spectra of naphthylisoquinoline alkaloids.[6] Indeed, again the theoretical CD spectrum of **2** matches very well with the experimental curve (Fig. 4), except for a systematically occurring shift of the spectrum by about 14 nm, which in Fig. 4 has been, for an easier comparison, subtracted optically by hatching. Also the comparison of the theoretical spectrum of **2** with the experimental one of the dimeric structures of the michellamines (see Fig. 2) fully confirms our structural attribution for these fascinating quateraryls.

3. Predicted Circular Dichroic Behavior of Michellamine 'Monomers':

Encouraged by this good agreement between theoretical and experimental CD spectra for ancistrobrevine B (**2**), we have then approached, step by step, the molecular halves of the natural michellamines, with more and more free phenolic OH-groups. Fig. 5 (upper part) shows the structure **3** of a hypothetical naphthylisoquinoline alkaloid, which has identical stereochemical features as michellamine A (**1a**), with only one extra methyl ether and **4** (Fig. 5, lower part) finally is the completely authentic monome-

ric unit. Indeed, the theoretically predicted CD spectra for these 'halves' now very much resemble the experimental spectrum of the existing dimer michellamine A (**1a**) (cf. Fig. 2).

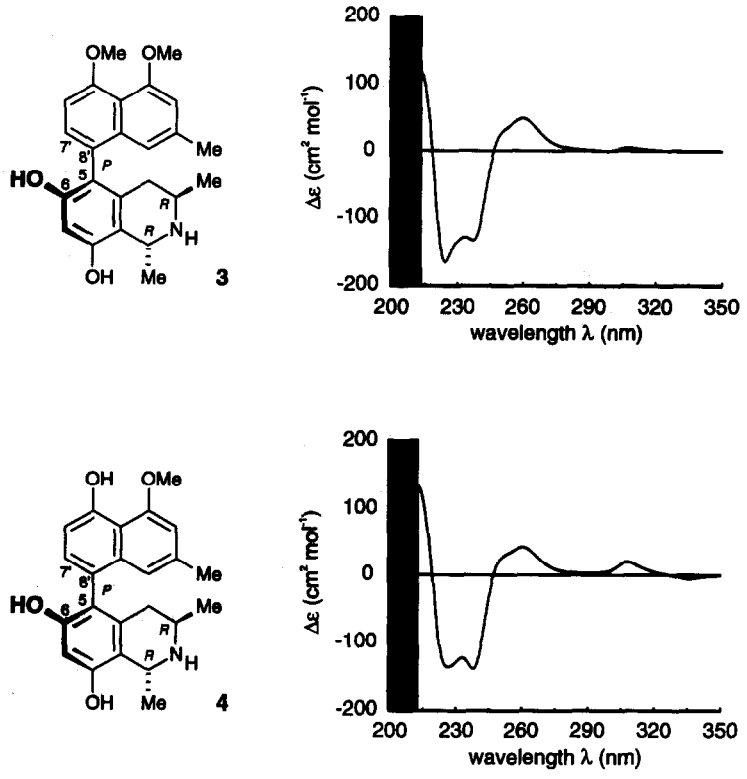


Fig. 5. Predicted (*i.e.* calculated) CD spectra of hypothetical monomers.

4. Conclusions and Further Perspectives:

In summary, the absolute stereostructure of the michellamines, as previously proposed[5] could now fully be confirmed by a comparison of their CD spectra with the experimental and the calculated ones of the structurally similar, yet 'monomeric' naturally occurring naphthylisoquinoline ancistrobrevine B (**2**), and with the computationally predicted spectra of the molecular 'halves', especially of structure **4**.

Still, the calculation of the CD spectra even of the whole, intact quateraryls **1**, despite their larger molecular sizes and their distinctly more important conformational flexibilities, remains a challenging goal, also in order to gain a better understanding and rationalization of the unusual effects at 250–340 nm. This work is in progress.

EXPERIMENTAL AND COMPUTATIONAL SECTION

Experimental:

The natural products ancistrobrevine B (**2**), michellamine A (**1a**), B (**1b**), and C (**1c**) were isolated and purified using procedures described earlier.[2,3] CD and UV spectra of these alkaloids were recorded in ethanolic solution on a Jobin Yvon Model CD6 spectrograph at room temperature within the range of 200–350 nm. Selected UV data of the michellamines: UV λ_{max} (ε): 228.0 (71915), 237.5 (65480), 260.5 (35957). For further spectroscopic data of michellamines A – C, see ref. 5.

Computational:

The computational generation of the theoretical CD spectra essentially followed a procedure elaborated earlier for the related naphthylisoquinoline alkaloid ancistrocladine,[6] although the alkaloids discussed in this article have only 3 *ortho*–substituents next to the axis (*i.e.* one less than ancistrocladine) and therefore are conformationally more flexible at the stereogenic axis. The calculation involved 3 steps:

1. First of all, the global energetic minimum structure of the corresponding molecule was calculated, from which the optimum dihedral angle $\sphericalangle\vartheta$ at the biaryl axis was obtained (for ϑ values of **2**, **3**, and **4**, see Table 1). Starting from this structure, this dihedral angle as a fixed geometry parameter was varied by small amounts (ca. 2–5°) and for each such given angle, the rest of the molecule was energetically optimized. By this procedure, a potential curve was constructed reflecting the energy of the molecule as a function of the axial distortion.

Tab. 1. Calculated angles (AM1[7]) for the minimum structures of ancistrobrevine B (**2**), the hypothetic 'molecular half' **4** of michellamine A (**1a**), and its methyl ether **3**.

dihedral angle $\sphericalangle\vartheta$ 7′,8′,5,6		
ancistrobrevine B **2**	structure **3**	structure **4**
79.7°	–80.0°	–81.7°

2. For 40 such structures lying on this potential curve, the rotational strength values R_{0a} for the electric transitions $0 \rightarrow a$ were calculated according to the dipol velocity formalism, which, in contrast to the formula primarily derived by Rosenfeld,[8] delivers origin–independent results even for approximated wave functions ψ_0 and ψ_a:

$$R_{0a} = \mathrm{Im}\{\frac{e\hbar}{im(E_a - E_0)} < \psi_0|\vec{p}|\psi_a > \cdot < \psi_a|\vec{m}|\psi_0 >\}.$$

The wave functions ψ_0 and ψ_a were obtained by a CNDO/2S–CI calculation,[9,10] where the CI expansion consists of the ground state determinant and 100 singly occupied configurations.

In order to get $\Delta\varepsilon$ curves from the calculated rotational strength values, we used the following formula:

$$\Delta\varepsilon(\lambda) = \sum_a \frac{16\pi^2\lambda N_A R_{0a}\sigma_{0a}(\lambda)}{3(2303)\hbar c},$$

where N_A denotes the Avogadro's number and $\sigma_{0a}(\lambda)$ a Gaussian band shape function.[11]

3. The single spectra thus obtained where then added up to the predicted overall spectrum by a Boltzmann–weighted averaging according to the calculated energies of the corresponding geometries. As in similar cases before,[6] the calculated $\Delta\varepsilon$ values had to be scaled by an empirical factor of $f = 2.8$, pointing at a systematic mistake within the formalism applied for the calculation of the rotational strengths.

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