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Cyclomegistine, the first alkaloid with the new cyclobuta[*b*]quinoline ring system from *Sarcomelicope megistophylla*

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Abstract—A new quinolone, cyclomegistine, was isolated from the bark of *Sarcomelicope megistophylla*. This alkaloid possesses the cyclobuta[*b*]quinoline ring system that has not been previously described either from natural or from synthetic origin. Biogenetically, cyclomegistine could arise from the oxidative aromatic ring cleavage of an acridone precursor, followed by photoisomerization of the resulting butadiene into the isomeric cyclobutene.

Sarcomelicope megistophylla Hartley (Rutaceae) is a small to medium sized tree, 8–12 m high, easily recognized by its pubescent leaves, exceptionally large for the genus (up to 35 cm long). This species is endemic to the region of Néaoua, New Caledonia.[1] Recently, we described the chemical constituents of its leaves[2] and the major alkaloids of the bark.[3] In a continuation of our studies of the genus *Sarcomelicope*,[4] we report here the isolation[5] and structure determination by means of spectral data (1D and 2D NMR,[6] MS,[7] X-ray[8]) of a new alkaloid, cyclomegistine (**1**) from the bark of *S. megistophylla*.

Cyclomegistine (**1**), was obtained as an optically inactive yellow product and its molecular formula was determined by HRMS as $C_{18}H_{19}NO_7$ [HR-EIMS m/z $[M]^+$ 361.1157, calcd 361.1162]. The UV spectrum recorded in MeOH was suggestive of a quinolone derivative. A typical hypsochromic shift observed upon addition of acid gave evidence for a 4-quinolone basic skeleton. In the aromatic region, the ^{1}H NMR spectrum displayed the characteristic signals associated with the four aromatic protons of the A ring of a 4-quinolone. At higher field, typical signals accounted for four OMe groups and one NMe group. The ^{13}C NMR spectrum showed three carbonyl resonances at 172.1, 169.1 and 166.8 ppm, respectively. The first confirmed the presence of the quinolone system and the others were assigned to two carbomethoxy groups. Additionally, the signals of two aliphatic quaternary oxygenated carbons were observed at 90.2 and 88.6 ppm. Further information on the structure of **1** was obtained from the long-range C–H correlations in the HMBC spectrum (Fig. 1). Three bond correlation between H-5

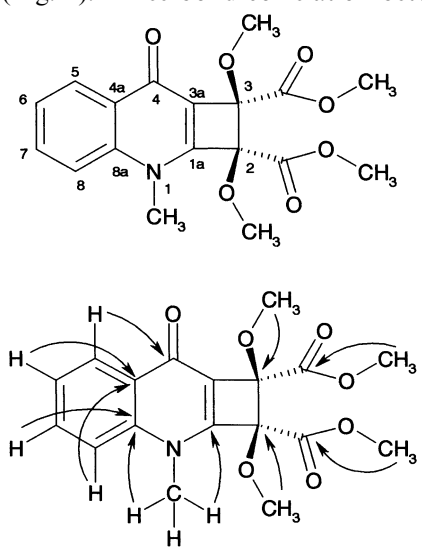

Figure 1. The structure of **1** and selected HMBC correlations.

Keywords: *Sarcomelicope megistophylla*; alkaloids; quinolone; Rutaceae; cyclobutene.

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and the carbonyl carbon C-4 confirmed the 4-quinolone ring system. The ^3J correlations of the NMe protons led to the identification of the two quaternary aromatic carbons 1a and 8a. Carbon 4a was identified by its ^3J correlations with H-6 and H-8. The last carbon of the quinolone ring was C-3a, which was observed at 121.01 ppm. Each of the two aliphatic quaternary oxygenated carbons C-1′ and C-2′ was correlated with an OMe group. Apparently, these two oxygenated quaternary carbons also carry the two carbomethoxy groups. Based on the molecular weight, it was also obvious that C-2 and C-3 were connected with a single bond forming a cyclobutane ring.

The cyclobutane moiety of the structure could not be confirmed by any NMR technique but only with X-ray crystallography. Crystallization of cyclomegistine from acetone led to pale yellow crystals of sufficient quality for the X-ray structure determination (Fig. 2).

It is noteworthy that the new alkaloid possesses the cyclobuta[*b*]quinoline ring system that has not been previously described either from natural or from synthetic origins. Additionally, the new compound contains in its structure the 1,2-dimethoxy-1,2-dicarbomethoxy-cyclobutene moiety, which is reported herein for the first time. When evaluated for its cytotoxic activity against L1210 leukemia cells,[9] cyclomegistine showed a moderate activity ($IC_{50}=80$ μM).

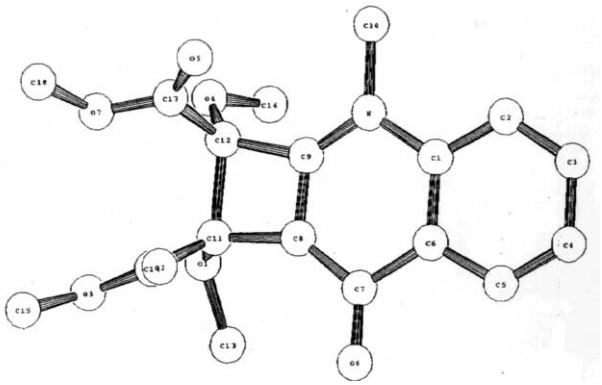
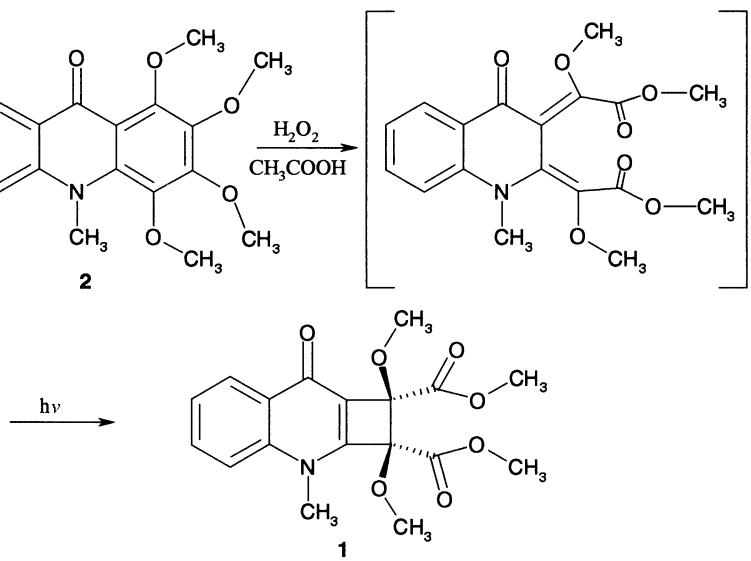


Figure 2. ORTEP drawing of **1**.

From a biogenetic point of view, cyclomegistine could be considered to be derived from a 1,2,3,4-tetraoxygenated acridone, such as melicopicine (**2**), which is one of the major alkaloids of *S. megistophylla* bark. A reasonable biosynthetic pathway would involve an oxidative cleavage of the C-2, C-3 double bond of the aromatic A-ring, facilitated by the presence of the four electron-donating groups,[10] followed by an electrocyclic photochemical ring closure of the 1,3-diene into the corresponding cyclobutene,[11] as outlined in Scheme 1. Indeed, the racemic character of natural cyclomegistine is in good agreement with a non enzymatic formation of the cyclobutene ring. In addition, the relative *cis* configuration of the four-membered ring substituents is identical with the stereochemistry observed for cyclobutenes resulting from the photoisomerization of 1,4-disubstituted 1,3-butadienes.[12]

The above mentioned biosynthetic pathway was confirmed by a biomimetic synthesis of cyclomegistine.[13] Oxidation of melicopicine (**2**)[14] with H_2O_2 in glacial acetic acid and simultaneous irrradiation of the reaction mixture led to a mixture from which a product with identical physical and spectral data to cyclomegistine was isolated in low yield.

The possibility of the artificial origin of **1** should be excluded, due to the co-occurrence of numerous oxidized acridone derivatives[3] in the plant, which indicates the existence of a common oxidative pathway.



Scheme 1. Biomimetic synthesis of cyclomegistine.

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References

1. Hartley, T. G. *Adansonia* **1986**, *8*, 183–189.
2. (a) Skaltsounis, A. L.; Seddrati, L.; Tillequin, F.; Koch, M.; Pusset, J.; Sévenet, T. *Nat. Prod. Lett.* **1995**, *5*, 281–287; (b) Fokialakis, N.; Mitaku, S.; Mikros, E.; Skaltsounis, A. L.; Tillequin, F.; Sévenet, T. *Phytochemistry* **1999**, *52*, 1745–1748.
3. (a) Papageorgiou, M.; Fokialakis, N.; Mitaku, S.; Skaltsounis, A. L.; Tillequin, F.; Sévenet, T. *J. Nat. Prod.* **2000**, *63*, 385–386; (b) Fokialakis, N.; Magiatis, P.; Skaltsounis, A. L.; Tillequin, F.; Sévenet, T. *J. Nat. Prod.* **2000**, *63*, 1004–1005; (c) Fokialakis, N.; Magiatis, P.; Skaltsounis, A. L.; Tillequin, F.; Sévenet, T. *Z. Naturforsch.* **2000**, *55C*, 874–876; (d) Fokialakis, N.; Magiatis, P.; Mitaku, S.; Tillequin, F.; Sévenet, T. *Chem. Pharm. Bull.* **2000**, *48*, 2009–2010.
4. (a) Mitaku, S.; Skaltsounis, A. L.; Tillequin, F.; Koch, M.; Pusset, J.; Chauvière, G. *J. Nat. Prod.* **1986**, *49*, 1091–1095; (b) Mitaku, S.; Skaltsounis, A. L.; Tillequin, F.; Koch, M.; Pusset, J.; Chauvière, G. *Heterocycles* **1987**, *26*, 2057–2063; (c) Mitaku, S.; Skaltsounis, A. L.; Tillequin, F.; Koch, M.; Pusset, J. *Ann. Pharm. Fr.* **1989**, *47*, 149–156; (d) Mitaku, S.; Skaltsounis, A. L.; Tillequin, F.; Koch, M.; Pusset, J.; Sévenet, T. *Nat. Prod. Lett.* **1995**, *7*, 219–225; (e) Tillequin, F. *Rec. Res. Dev. Phytochem.* **1997**, *1*, 675–687.
5. Isolation: Plant material, extraction and purification as described in Ref. 3a. Fraction 5 was chromatographed with MPLC (cHex, EtOAc gradient) to afford compound **1** (8 mg).
6. Spectral data for compound **1**: ^{1}H NMR ($CDCl_3$/TMS, 400 MHz, δ ppm, J in Hz): 3.69 (3H, s, OCH_3-2), 3.70 (3H, s, OCH_3-3), 3.74 (3H, s, NCH_3), 3.75 (3H, s, $COOCH_3$-2), 3.77 (3H, s, $COOCH_3$-3), 7.45 (1H, td, J=8.8, 1.5 Hz, H-6), 7.48 (1H, dd, J=8.8, 1.5 Hz, H-8), 7.73 (1H, td, J=8.8, 1.5 Hz, H-7), 8.49 (1H, dd, J=8.8, 1.5 Hz H-5), ^{13}C NMR ($CDCl_3$/TMS, 50 MHz, δ ppm): 172.11 (C-4), 169.12 ($\underline{C}OOCH_3$-2), 166.79 ($\underline{C}OOCH_3$-3), 152.97 (C-1a), 142.09 (C-8a), 132.42 (C-7), 129.94 (C-4a), 127.62 (C-5), 124.22 (C-6), 121.01 (C-3a), 115.33 (C-8), 90.21 (C-2), 88.64 (C-3), 56.58 (OCH_3-2), 56.34 (OCH_3-3), 52.97 ($COOCH_3$-2), 52.72 ($COOCH_3$-3), 34.82 (NCH_3); $[\alpha]_D^{25}=0°$ (c 0.07, CH_2Cl_2); UV (MeOH): λ_{max} (log ε) 332 (2.52), 318 (2.46) nm.
7. MS-DCI m/z: 362 $[M+H]^+$; EI m/z (rel. int): 361 (35), 346 (80), 333 (30), 318 (100), 302 (70), 272 (40), 244 (45).
8. Crystallographic data (excluding structure factors) for the structures in this paper have been deposited with the Cambridge Crystallographic Data Centre as supplementary publication number CCDC 161469; mp 146°C.
9. The murine leukemia was from the American Type Culture Collection (Rockville Pike, MD). Cells were grown in RPMI medium supplemented with 10% fetal calf serum, 2 mM L-glutamine, 100 U/ml penicillin, 100 mg/ml streptomycin and 10 mM HEPES buffer (pH 7.4). The cytotoxicity was measured by the microculture tetrazolium assay.
10. For relevant examples of oxidative cleavage of aromatic rings, see: (a) Sucharda, E. *Ber. Dtsch. Chem. Ges.* **1925**, *58*, 1727–1729; (b) Bösenken, J.; Sloof, G. *Recl. Trav. Chim. Pays-Bas* **1930**, *49*, 100–101; (c) Tsuji, J.; Takayanagi, H. *Tetrahedron* **1978**, *34*, 641–644; (d) Bankston, D. *Org. Synth.* **1987**, *66*, 180–183.
11. For a discussion on the photochemical rearrangement of butadienes into cyclobutenes, see: Laarhoven, W. H. *Org. Photochem.* **1987**, *9*, 129–224.
12. Srinivasan, R. *J. Am. Chem. Soc.* **1968**, *90*, 4498–4499.
13. To a solution of melicopicine (**2**) (30 mg, 0.09 mmol) in glacial acetic acid (1.5 mL) was added H_2O_2 (30%, 1.5 mL) and the reaction mixture was refluxed and irradiated with a Pyrex-jacketed lamp for 6 h. Then the reaction mixture was neutralized with $NaHCO_3$ and extracted with CH_2Cl_2/H_2O. The organic phase was collected and evaporated. The residue was purified with preparative TLC (CH_2Cl_2–MeOH 99:1) to afford **1** (2 mg, 6%).
14. Melicopicine (**2**) used as starting material for the synthesis of **1** was isolated from the bark of *S. megistophylla* as described in Ref. 3a.