Megistolactone, a New Alkaloid from *Sarcomelicope megistophylla*

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A new quinolone alkaloid, megistolactone (**1**) was isolated from the bark of *Sarcomelicope megistophylla.* Its structure has been elucidated on the basis of MS and NMR data. From a biogenetic point of view, this compound should be considered as an oxidation product of 1,2,3,4-tetra-*O*-subsituted acridone alkaloids, which are also present in the bark.

Introduction

Sarcomelicope megistophylla Hartley (Rutaceae) is an 8–12 m high tree, easily recognized by its pubescent leaves, exceptionally large for the genus (up to 35 cm long). This species is endemic to the region of Nouméa, New Caledonia (Hartley, 1986). Recently, we described the chemical constituents of its leaves (Skaltsounis *et al.*, 1995; Fokialakis *et al.*, 1999) and the major alkaloids of the bark (Papageorgiou *et al.*, 2000; Fokialakis *et al.*, 2000). In a continuation of our studies of the genus *Sarcomelicope* (Tillequin, 1997), we report here the isolation and structure determination of a novel 4-quinolone alkaloid, from the bark of *Sarcomelicope megistophylla.*

Results and Discussion

Megistolactone (**1**) was obtained as a yellowish amorphous product. The molecular formula was determined by HRMS as $C_{13}H_{11}NO_4$. 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 (Rapoport and Holden, 1960; Hā-huy-Kê *et al.*, 1970). The IR spectrum showed two characteristic bands at 1647 cm^{-1} and 1789 cm^{-1}. The first one corresponds to the carbonyl group of the quinolone and the second one to the carbonyl group of a 5-membered lactone ring. In the aromatic region, the ^1H 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 one *O*Me group and one *N*Me group, whereas the signal of one deshielded methine proton appeared as a singlet at 6.15 ppm. The ^{13}C NMR spectrum showed two carbonyl resonances, at 172.3 and 164.8 ppm. The former confirmed the presence of the quinolone system and the latter was assigned to an unsaturated lactone. Additionally, the signal of a methine carbon joined with two oxygen atoms was observed at 96.8 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 correlations between the methine proton at δ 6.15 and the *O*Me carbon at 56.6 ppm on one hand, and the carbonyl group at 164.8 ppm on the other hand, indicated that the aliphatic methine carbon beared the methoxy group and was also included in the

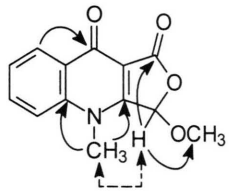

Fig. 1. Selected HMBC (——) and NOESY (-----) correlations for megistolactone (**1**).

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lactone ring. Correlation between the three-proton singlet of the *N*Me group and the two quarternary carbon signals of C-2, at 160.3 ppm, and C-10, at 140.9 ppm, gave incidentally additional evidence for the 4-quinolone basic skeleton. Finally, determination of the structure of the new natural product as **1**, and discrimination against the alternative structure **2** (Fig. 2), was provided by a NOESY experiment, which showed a strong cross peak between the signals of the *N*Me and the aliphatic methine proton. The absolute configuration of the chiral center at C-11 could not be determined, due to the small amount of the product isolated. When evaluated for its cytotoxic activity against L1210 leukemia cells, megistolactone only showed a moderate activity (IC_{50} = 70 μM).

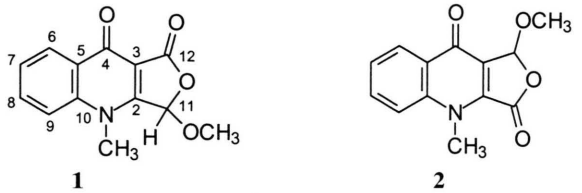

Fig. 2. The structure of megistolactone (**1**) and the alternative structure **2**.

Biogenetically, megistolactone (**1**) should be considered as resulting from the oxidation of the A aromatic ring of an acridone, such as melicopine, melicopidine, or melicopicine which are the major alkaloids of the bark. The presence of four electron-donating groups on the A ring of these 1,2,3,4-tetra-*O*-substituted acridones should greatly facilitate their oxidative cleavage. From a chemotaxonomic point of view, it is interesting to note that a similar biogenesis, involving the oxidative ring opening of the aromatic ring of an acridone, had been postulated for 1-methyl-4-quinolone-2,3-dicarboxylic acids dimethylester, previously isolated from *Sarcomelicope dogniensis* (Mitaku *et al.*, 1995).

Experimental

General experimental procedures

Optical rotation was measured with a Perkin-Elmer 341 polarimeter. UV spectra were recorded on a Shimadzu-160A spectrophotometer. The IR spectrum was obtained on a Perkin-Elmer Paragon 500 instrument. NMR spectra were recorded on Bruker DRX 400 and Bruker AC 200 spectrometers [^{1}H (400 and 200 MHz) and ^{13}C (50 MHz)]; chemical shifts are expressed in ppm downfield to TMS. The 2D NMR experiments were performed using standard Bruker microprograms. EIMS and HRMS were determined on HP-6890 and AEI MS-902 spectrometers, respectively.

Plant material

The plant material was collected at Nouméa (New Caledonia) in May 1984. Herbarium samples (Pusset-Chauvière 261) are deposited in the herbarium of the Centre ORSTOM at Nouméa (New Caledonia).

Extraction and isolation

Extraction of alkaloids as decribed (Papageorgiou *et al.*, 2000). The dichloromethane bark extract was chromatographed over a CC silica gel Merck 0.04–0.06 mm (flash), using cyclohexane-EtOAc gradient to give 6 fractions. Fraction 5 was rechromatographed to afford megistolactone **1** (10 mg).

Spectroscopic data

Megistolactone (**1**), $[\alpha]_D$ −3° (0.1 g/100 ml, CH_2Cl_2); UV (MeOH) λ_{max} (log ε) 245 (3.88), 252 (sh), 299 (sh), 310 (3.75), 323 (3.69) nm; IR (CH_2Cl_2) ν_{max} 2951, 2928, 1789, 1647, 1261, 1207 cm^{-1}; ^{1}H NMR ($CDCl_3$, 400 MHz, δ ppm): 3.85 (3H, s, N-CH_3), 8.48 (1H, dd, J = 8, J = 1.5 Hz, H-6), 7.48 (1H, t, J = 8 Hz, H-7), 7.76 (1H, td, J = 8, J = 1.5 Hz, H-8), 7.56 (1H, d, J = 8, H-9), 6.15 (H, s, H-11) 3.64 (3H, s, CH_3O-11); ^{13}C NMR ($CDCl_3$, 50 MHz, δ ppm): 35.0 (N-CH_3), 160.3 (C-2), 104.4 (C-3), 172.3 (C-4), 128.9 (C-5), 127.5 (C-6), 125.8 (C-7), 133.6 (C-8), 115.9 (C-9), 140.9 (C-10), 96.8 (C-11), 56.6 (CH_3O-11), 164.8 (C-12); HRMS found: 245.0647 (calcd for $C_{13}H_{11}O_4N$, 245.0688); MS-DCI *m/z* 246 $[M+H]^+$.

Cytotoxicity assay

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 N-(2-hydroxyethyl)piperazine-N′-(2-ethanesulfonic acid) (HEPES) buffer (pH 7.4). The cytotoxicity was measured by the microculture tetrazolium assay (Pierré *et al.*, 1991).

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