Tetrahedron Letters, Vol.31, No.47, pp 6881-6882, 1990
Printed in Great Britain

0040-4039/90 $3.00 + .00
Pergamon Press plc

NEW TRITERPENE DIMERS FROM *MAYTENUS ILICIFOLIA*

Hideji Itokawa,[a,*] Osamu Shirota,[a] Hiroshi Morita,[a] Koichi Takeya,[a] Nobuo Tomioka[b] and Akiko Itai[b]
Tokyo College of Pharmacy,[a] Horinouchi 1432-1, Hachioji, Tokyo 192-03, Japan
Faculty of Pharmaceutical Sciences, The University of Tokyo,[b] Hongo, Bunkyo-ku, Tokyo 113, Japan

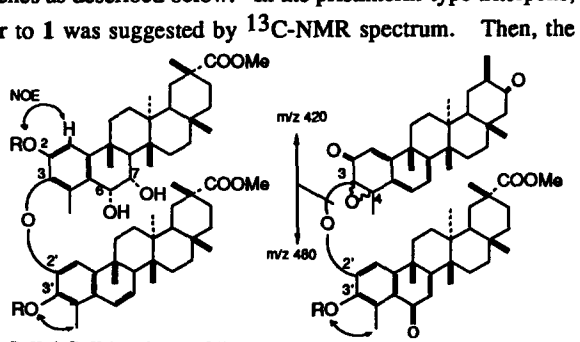
Abstract: Four new cytotoxic triterpene dimers from *Maytenus ilicifolia* were elucidated by spectroscopic and chemical evidences. Two of them were related to be atropisomer separated by a barrier of 32.8 kcal/mol and each stable conformer was elucidated by molecular mechanics calculation and NOE relationship.

Maytenus ilicifolia (Celastraceae) have been used for anticancer and contraception as medicinal folk plant named "Cangorosa" in South America[1] and the presence of maitenin and pristimerin as toxic principles so far have been revealed.[2] Purification of the methanol extract by the guidance of cytotoxic assay led us to isolate new triterpene dimers named as cangorosin A(**1**), atropcangorosin A(**2**), dihydroatropcangorosin A(**3**) and cangorosin B(**4**).[3] This paper is dealed with the structures of them and conformational analysis of **1** and **2** related to be atropisomer.

Cangorosin A(**1**)[4] showing the molecular formulae, $C_{60}H_{84}O_9$ by FAB-MS spectrum was suggested to be a triterpene dimer and was comprised of two pristimerin-type triterpenes. The presence of aromatized A ring containing a hydroxy and ether connected parts was presumed each other by the signal of ^{13}C-NMR spectrum.[5] One of the constituting triterpenes was found to possess a 1,2-glycol at C-6 and C-7 by the ABX spin system, whose configurations were α by the ^{1}H coupling constants (2.9 and 11.3 Hz)[6] among H-6, H-7 and H-8, respectively. The presence of double bond in the other triterpene was suggested by the ^{1}H-signals (δ 6.66 and 5.89).[6] The relationship as described abcve was also supported by ^{1}H-^{13}C COSY and COLOC spectra. The ether connected site of each other triterpene was determined by NOEs among the methoxy-methyl and aromatic protons in a dimethyl derivative (**5**) which was prepared by iodomethane as shown in Figure.

Atropcangorosin A(**2**)[7] was found to be the same structure to **1** by the similar procedure and appeared at 150°C in DMSO solution of **1** with an estimated half-life of 3 hr, indicating a barrier of 32.8 kcal/mol.[8] Dihydroatropcangorosin A(**3**)[9] was identical with the compound obtained by hydrogenation of **2** with H_2/PtO_2.

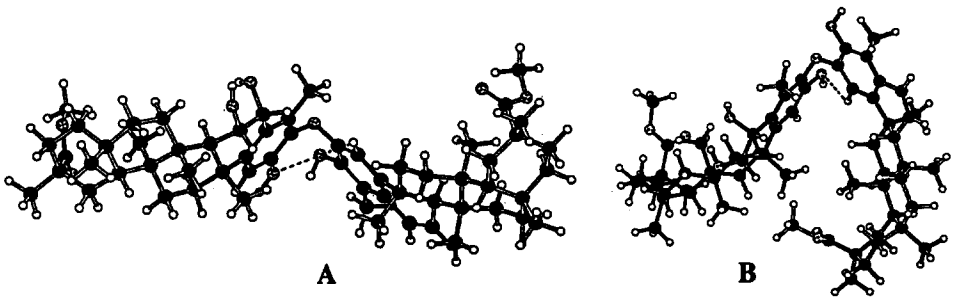
Cangorosin B(**4**)[10] showing the molecular formulae, $C_{58}H_{74}O_8$ by FAB-MS spectrum turned out to consist of a pristimerin- and tingenone-type triterpenes as described below. In the pristimerin-type triterpene, the presence of an aromatic A ring system similar to **1** was suggested by ^{13}C-NMR spectrum. Then, the presence of an unsaturated ketone moiety at B ring was suggested by the signals of δ126.43(d), 170.91(s) and 187.22(s) in ^{13}C-NMR spectrum. The other triterpene was considered to be tingenone type comparing with the data of tingenone.[11] The structure of A and B linkage was implied by UV absorption band at 378 nm and the NMR data, and was secured by ^{1}H-^{13}C long rang coupling system in COLOC spectrum. The presence of epoxide at C-3 and C-4 was



1: R=H; 2: R=H (atropisomer of 1)
3: 6', 7'-dihydro derivative of 2
5: R=CH_3 (derivative of 1); 6: R=CH_3 (derivative of 2)
4: R=H
7: R=CH_3

verified by the carbon chemical shifts at δ 92.01(s) and 79.53(s), and was not in conflict with fragmentation pattern (*m/z* 420 and 480) in EI-MS spectrum. The ether connected site of each other triterpene was determined by NOEs between the methoxy-methyl and aromatic proton in a methyl derivative (**7**) which was prepared by iodomethane.

Conformational relationship of **1** and **2** was studied by axis rotates per 5 degrees around ether oxygen connected at A ring each other.[12] Two of the most energetically stable conformers were further refined by MM2[13] to obtain conformers **A** and **B**(**A**: 132.3 kcal/mol, **B**: 133.6 kcal/mol). More stable **A** was considered to correspond to **1** and **B** to **2** whose conformer was consistent with the fact that the NOE between methoxyl-methyl at C-2 and an aromatic proton at C-1' in **6** was observed (Atomic distance from the oxygen at C-2 to hydrogen at C-1'; **A**: 4.39Å, **B**: 2.95Å).



References and Notes

1. C.M.O. Simoes, et al; "Plantas da medicina popular no Rio Grande do Sul", 1986, Ed. da Universidade/UFRGS.
2. a) O.G. de Lima, J.S. de B.Coelho, E. Weigert, I.L. D'Albuqerque, D. de A.Lima and M.A. de M.E Souza, Rev. Inst. Antibio. Recife, **11**, 35 (1971); b) F.D. Monache, G.B.M-Bettolo, O.G. de Lima, I.L. D'Albuquerque and J.S. de B. Coelho, Gazz. Chem. Itali., **102**, 317 (1972); c) M.S. Ahmed, H.H.S. Fong, D.D. Soejarto, R.H. Dobberstein, D.P. Waller and R.M-Azorero, J. chromatogr., **213**, 340 (1981).
3. Cytotoxic activity of **4**: IC_{50} 3.7×10^{-3} mol/l against p388 cells.
4. Cangorosin A(**1**): Colorless amorphous powder, $[\alpha]_D$ +237.4°(c 0.25, $CHCl_3$), FAB-MS(pos.+KI) *m/z* 969(M^+-18+K), 931(M^+-18+H), 930(M^+-18). EI-MS m/z(%): 466(7), 465(20, Calcd. for $C_{30}H_{41}O_4$: 465.3002; Found: 465.3007).
5. ^{13}C-NMR assignment of each A ring in **1**($CDCl_3$, 100MHz): 108.30(C-1), 144.41(C-2), 140.68(C-3), 123.17(C-4), 124.69(C-5), 143.66(C-10), 107.97(C-1'), 141.60(C-2'), 139.07(C-3'), 122.01(C-4'), 124.13(C-5'), 143.76(C-10').
6. ^{1}H-NMR ($CDCl_3$, 400MHz) **1**: 0.77(3H, s), 0.84(3H, s), 1.03(3H, s), 1.08(3H, s), 1.12(6H, s), 1.20(6H, s), 1.30(3H, s), 1.34(3H, s), 2.18(3H, s), 2.33(3H, s), 2.38(1H, d, J=11.3), 2.49(1H, t), 3.58(3H, s), 3.63(3H, s), 4.88(1H, d, J=2.9), 4.97(1H, dd, J=2.9, 11.3), 5.89(1H, dd, J=2.7, 10.0), 6.46(1H, s), 6.66(1H, dd, J=2.9, 10.0), 6.68(1H, s).
7. Atropcangorosin A(**2**): Colorless powder, $[\alpha]_D$ +76.4°(c 0.28, $CHCl_3$), FAB-MS (pos.) *m/z* 930(M^+-18), EI-MS *m/z*(%): 466(63, Calcd. for $C_{30}H_{42}O_4$: 466.3080; Found: 466.3040), 464(46, Calcd. for $C_{30}H_{40}O_4$: 464.2924; Found: 464.2907). ^{1}H-NMR($CDCl_3$, 400MHz): 0.79(3H, s), 0.88(3H, s), 0.98(3H, s), 1.07(3H, s), 1.12(3H, s), 1.19(3H, s), 1.20(3H, s), 1.32(3H, s), 1.35(3H, s), 2.16(3H, s), 2.34(3H, s), 2.39(1H, d, J=11.2), 2.51(1H, br.s), 3.58(3H, s), 3.66(3H, s), 4.87(1H, d, J=3.0), 5.06(1H, dd, J=3.0,11.2), 5.90(1H, dd, J=2.6, 9.9), 6.56(1H, s), 6.66(1H, dd, J=2.9,9.9), 6.68(1H, s).
8. H. Gunther, "NMR Spectroscopy"; Wiley: New York, 1980; pp 240-244.
9. Dihydroatropcangorosin A(**3**): Colorless powder, $[\alpha]_D$ +87.3°(c 0.45, $CHCl_3$), FAB-MS (pos.) *m/z* 932(M^+-18), EI-MS *m/z* (%):468(100, Calcd. for $C_{30}H_{44}O_4$: 468.3237; Found: 468.3253), 467(22, Calcd. for $C_{30}H_{43}O_4$:467.3159;Found:467.3179).
10. Cangorosin B(**4**): Yellow amorphous powder, $[\alpha]_D$ +483.3° (c 0.25, $CHCl_3$), FAB-MS (pos.) *m/z*: 899(M^++H); (pos.+KI) *m/z*: 937(M^++K), 899(M^++H). EI-MS m/z(%): 480 (54, Calcd. for $C_{30}H_{40}O_5$: 480.2873; Found: 480.2859), 465(22), 420(54, Calcd. for $C_{28}H_{36}O_3$: 420.2662; Found 420.2650). ^{1}H-NMR($CDCl_3$, 400MHz): 0.63(3H, s), 0.97(3H, s), 0.98(3H, d, J=6.4), 0.99(3H, s), 1.11(3H, s), 1.19(3H, s), 1.27(3H, s), 1.30(3H, s), 1.49(3H, s), 1.55(3H, s), 1.58(3H, s), 2.48(3H, s), 2.84(1H, d, J=14.3), 3.57(3H, s), 4.97(1H, s), 5.98(1H, d, J=6.5), 6.12(1H, d, J=1.4), 6.21(1H, s), 6.33(1H, dd, J=1.4, 6.5), 7.00(1H, s).
11. V. Krishnamoorthy, J.D. Ramanathan and T.R. Seshadri, Tetrahedron Lett., 1047 (1962).
12. The coordinates required was made from pristimerol in the XDC database (biographic and numerical files from cambridge crystallographic data centre) and refined by PM3 in MOPAC ver 5.0. Total energy was evaluated by the sum of nonbonded interaction and electrostatic interaction in Scheraga 12-6 potential.
13. N.L. Allinger, MM2 (87).

(Received in Japan 10 September 1990)