CHEMISTRY LETTERS, pp. 1577-1580, 1980. © The Chemical Society of Japan 1980

ALBANINS F AND G, NATURAL DIELS-ALDER ADDUCTS FROM MULBERRY[1)]

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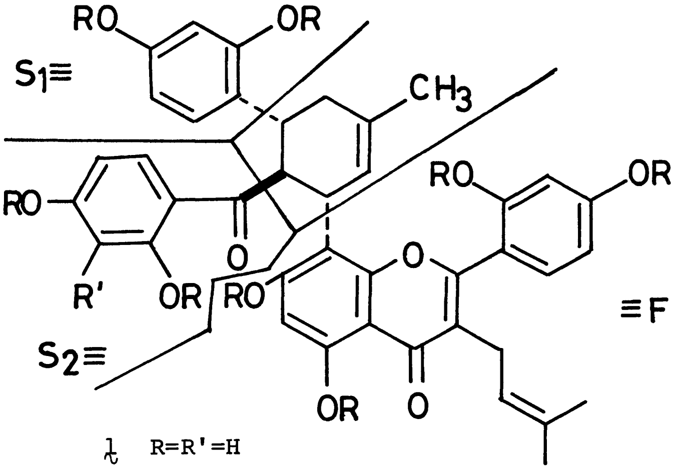
The structure elucidation of albanins F and G, two of the pre-existing antifungal principles of shoot epidermis of mulberry, is described.

In connection with our continuing studies on mulberry phytoalexins,[1)] we already reported the structures of several compounds,[2)] albanins A ∿ E, as antifungal principles[3)] pre-existing in shoot epidermis of mulberry (*Morus alba* Linné). Column chromatography over silica gel (CH_2Cl_2-CH_3OH) of acetone extracts of the shoot epidermis[2)] followed by rechromatography of the resulting polar fractions over Sephadex LH-20 led to isolation of two compounds, designated as albanins F (**1**) and G (**1b**), in 0.4 and 0.3% yields, respectively, from the dried epidermis. These compounds completely inhibited spore germination of *Bipolaris leersiae* at concentrations of 10^{-4} ∿ 10^{-5} M. We report herein the structure elucidation of albanins F and G.

Albanin F (**1**), $C_{40}H_{36}O_{11}$ (m/e 692.2248, M^+, FD-MS), amorphous and $[\alpha]_D$ -529° (CH_3OH), exhibited absorption maxima at 208 nm (δ 65000), 227 (sh, 34600), 265 (29300), 279 (sh, 19300), and 319 (14300), and at 1660 and 1620 cm^{-1} in the UV (C_2H_5OH) and IR (KBr) spectra, respectively. The ^{1}H-NMR spectrum (CD_3COCD_3) of **1** revealed the presence of eight phenolic hydroxy protons (ε 7.8 ∿ 13.0), ten aromatic protons (δ 5.9 ∿ 7.4), nine protons due to a prenyl group [δ 1.48 and 1.61 (each 3H, br s), 3.18 (2H, br d, J = 7 Hz), and 5.20 (1H, br t, J = 7)], one methyl protons [1.52 3H, br s)], and six other protons [2.0 (2H, m), 3.7 (1H, m), 4.43 (1H, br d, J = 11), 4.67 (1H, br t, J = 11), and 5.22 (1H, br s)]. Treatment of **1** with dimethyl sulfate and potassium carbonate in the presence of dicyclohexyl-18-crown-6 (in acetone, reflux, 19 h) effected exhaustive methylation to give its octamethyl ether (**1a**), amorphous and $[\alpha]_D$ -309° ($CHCl_3$); m/e 804 (16%, M^+, $C_{48}H_{52}O_{11}$, EI-MS), 761 (1, $C_{45}H_{45}O_{11}$), 639 (1), 476 (2, $C_{29}H_{32}O_6$), 328 (9, $C_{19}H_{20}O_5$), and 165 (100).

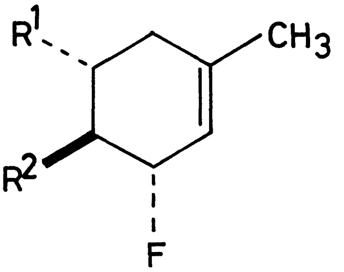
Pyrolysis (in toluene, 280 °C, 2 h) of the methyl ether (**1a**) in a sealed tube afforded two fragmentation products (**2a** and **3a**) in 54 and 37% yields, respectively, after preparative TLC. The former (**2a**), $C_{19}H_{20}O_5$ (m/e 328.1293, M^+), mp 128-129 °C, showing the ^{1}H-NMR signals at δ ($CDCl_3$) 3.86 and 3.91 (each 3H, s), 3.89 (6H, 1H, J = 8), was identified as *trans*-2,2',4,4'-tetramethoxychalcone by direct comparison with an authentic sample prepared by condensation of

2,4-dimethoxyacetophenone with 2,4-dimethoxybenzaldehyde. The latter (3a), $C_{29}H_{32}O_6$ (m/e 476.2179), gum, was presumed to be a flavone containing a cojugate diene moiety and a prenyl group from the ^{1}H-NMR spectrum: a diene moiety [δ (CDCl$_3$) 1.91 (3H, br s), 4.88 and 4.96 (each 1H, br s), 6.80 and 7.26 (each 1H, d, J = 16)]; a prenyl group [1.44 and 1.61 (each 3H, br s), 3.09 (2H, br d, J = 7), and 5.24 (1H, br t, J = 7)]; four methoxy protons; four aromatic protons [6.43 (1H, s), 6.56 (1H, d, J = 2), 6.57 (1H, dd, J = 9 and 2), and 7.37 (1H, d, J = 9)]. Indeed, the diene (3a) formed its hexahydro derivative on catalytic hydrogenation (Pt), which was identified as tetrahydrokuwanon C tetramethyl ether[4] (3b) by direct comparison with an authentic specimen. Disposition of the prenyl group at C-3 (not C-8) was deduced from comparison of the chemical shifts of the group protons with the corresponding shifts of kuwanon C tetramethyl ether [C-3 prenyl, δ (CDCl$_3$) 1.41 and 1.59 (each 3H, br s), 3.02 (2H, br d, J = 7), and 5.13 (1H, br t, J = 7); C-8 prenyl, 1.54 and 1.61 (each 3H, br s), 3.34 (2H, br d, J = 7), and 5.20 (1H, br t, J = 7)] and morusin trimethyl ether[5] [C-3 prenyl, δ (CDCl$_3$) 1.41 and 1.59 (each 3H, br s), 3.02 (2H, br d, J = 7), and 5.18 (1H, br t, J = 7)].



1 R=R'=H

1b R=H, R'=prenyl



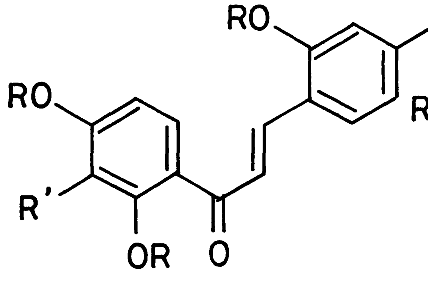
1 R^1=S_1, R^2=S_2, R=R'=H

1' R^1=S_2, R^2=S_1, R=R'=H

1a R^1=S_1, R^2=S_2, R=CH_3, R'=H

1b R^1=S_1, R^2=S_2, R=H, R'=prenyl

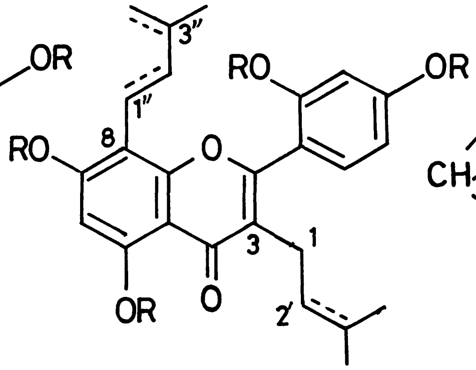
1c R^1=S_1, R^2=S_2, R=CH_3, R'=prenyl



2 R=H, R'=H

2a R=CH_3, R'=H

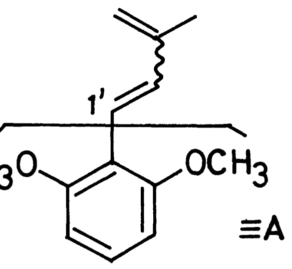
2b R=CH_3, R'=prenyl



3 R=H, $\Delta^{2',1'',3''}$

3a R=CH_3, $\Delta^{2',1'',3''}$

3b R=CH_3, no Δ



4 $\Delta^{1'}$ trans

5 $\Delta^{1'}$ cis

The formation of two fragmentation products (2a and 3a) strongly suggested that albanin F would be formulated as a Diels-Alder adduct of 2 and 3 and hence represented by either one of two possible formulas (**1** and **1**'). Detailed spin-decoupling experiments elucidated the presence of an expected cyclohexene moiety with signal patterns (δ and Hz) as shown in Fig. 1 (**1a**), in which the coupling constants between adjacent protons on the ring indicated that the cyclohexene ring would probably adopt a half-chair conformation with all the three substituents (R^1, R^2, and F) equatorial, and hence with two pairs of the trans-oriented adjacent substituents. The validity of formula **1** for albanin F was achieved by examination of ^{1}H-NMR spectra of the following pertinent model compounds.

Treatment of an ylide prepared from methallyltriphenylphosphonium chloride with 2,6-dimethoxybenzaldehyde afforded a 3:2 mixture of trans- and cis-dienes (**4** and **5**), which on heating with the trans-chalcone (**2a**) (in toluene, 160 °C, 61 h) formed a 2:1 mixture of two adducts (**6a** and **6b**) as major isolable products in a 36% yield. The mixture was separated by repeated column chromatography to give each adduct in pure state: **6a** (major), $C_{32}H_{36}O_7$ (m/e 532.2470, M^+), mp 159-161 °C; m/e 532 (17%), 367 (4), 204 (2), and 165 (100); λ_{max} (C_2H_5OH) 210 nm (ε 70900), 226 (sh 39900), 271 (16600), and 303 (10100); ν_{max} (KBr) 1665 and 1603 cm^{-1}; **6b** (minor), $C_{32}H_{36}O_7$ (m/e 532.2486, M^+), mp 163-165 °C; m/e 532 (3%), 367 (1), 204 (30), and 165 (100); λ_{max} 208 nm (ε 73800), 225 (sh, 37400), 269 (16100), and 300 (10000); ν_{max} 1670 and 1603 cm^{-1}. In view of the stereospecificity and regioselectivity due to substituents of Diels-Alder reactions,$^{6)}$ formulas **6a** and **6b**, differing only in (relative) configuration of a single carbon atom, were reasonably assigned to the major and minor adducts, respectively. The spectral data were also in good accord with the formulas. The ^{1}H-NMR signal patterns [Fig. 2 (**6a**) and 3 (**6b**)] due to protons on the relevant cyclohexene ring, obtained by spin-decoupling studies, clearly indicated that the methine proton [H_D, δ 4.82 (**6a**) and 4.7 (**6b**)] adjacent to the dimethoxybenzoyl group (S_2) appeared at lower fields than that [H_C, δ 3.7 (**6a**) and 4.24 (**6b**)] in the respective spectrum of the adducts. These chemical shifts, coupled with those [δ 4.61 (H_D) and 4.25 (H_C)] of chalcomoracin heptamethyl ether,$^{1)}$ elucidate that formula **1** with the indicated (relative) configuration is assigned most reasonably to albanin F.$^{7)}$

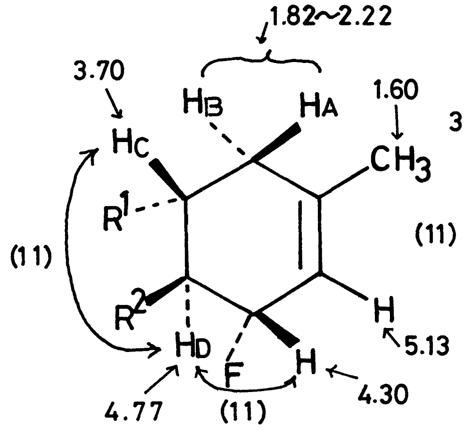


Fig. 1 (**1a**)

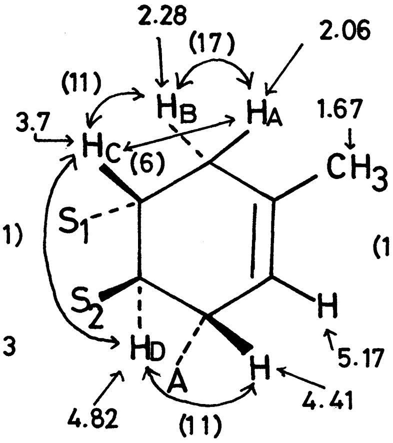


Fig. 2 (**6a**)

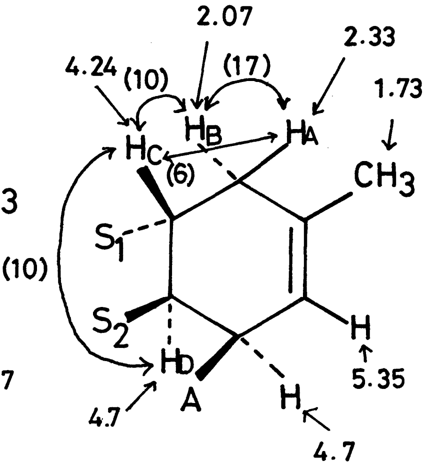


Fig. 3 (**6b**)

Albanin G (**1b**), amorphous, had a molecular formula of $C_{45}H_{44}O_{11}$ (m/e 760.2978, FD-MS) and $[\alpha]_D$ -455° (CH_3OH) and formed its octamethyl ether (**1c**), $C_{53}H_{60}O_{11}$ (m/e 872.3974), amorphous and $[\alpha]_D$ -335° ($CHCl_3$), which on pyrolysis gave tetramethylmorachalcone A1) (**2b**) and the diene (**3a**). Albanin G was then assigned formula **1b**, prenylalbanin F, in essentially the same manner as albanin F.

Albanins F (**1**) and G (**1b**) are considered to be formed by Diels-Alder type of reactions *in vivo*, because the compounds are optically active like chalcomoracin1) found in diseased mulberry.

References and Notes

1) Part 8 in the series "Studies on Phytoalexins of the Moraceae;" Part 7, M. Takasugi, S. Nagao, T. Masamune, A. Shirata, and K. Takahashi, Chem. Lett. (the preceding paper).

2) a) M. Takasugi, S. Nagao, L. Muñoz, s, Ishikawa, T. Masamune, A. Shirata, and K. Takahashi, 22nd Symposium on the Chemistry of Natural Products, Fukuoka, October 1979, Symposium Papers p. 275: albanins A, B, and C correspond to compounds **20**, **19**, and **21**, respectively. b) M. Takasugi, S. Ishikawa, T. Masamune, A. Shirata, and K. Takahashi, 42nd Annual Meeting of the Chemical Society of Japan, Sendai, September 1980, Abstr. Papers p. 352.

3) A. Shirata and K. Takahashi, Bull. Sericul. Exp. Sta., **27**, 637 (1978).

4) T. Nomura, Y. Sawaura, T. Fukai, S. Yamada, and S. Tamura, Heterocycles, **9**, 1355 (1978).

5) T. Nomura, T. Fukai, S. Yamada, and M. Katayanagi, Chem. Pharm. Bull., **26**, 1394 (1978).

6) J. Sauer, Angew. Chem. Internat. Edit., **6**, 16 (1967).

7) After completion of our work, Prof. Taro Nomura, Toho University, informed us that his group assigned the same *planar* formula to a compound named Kuwanon G [a private communication, June 14th and August 18th, 1980; T. Nomura and T. Fukai, Chem. Pharm. Bull., **28**, 2548 (1980)]. Kuwanon G and its related compound named kuwanon H were indistinguishable (only TLC and ^{1}H-NMR) from our compounds, albanins F and G, respectively. On the other hand, Prof. Hiroshi Hikino, Tohoku University, reported the structure of a hypotensive compound isolated from mulberry root barks and named moracenin B. The compound seems to be identical with albanin F (= kuwanon G) but the assigned (planar) formula (**1'**) is different from that (**1**) of our compound [Y. Oshima, C. Konno, H. Hikino, and K. Matsushita, Tetrahedron Lett., **21**, 3381 (1980)]. If moracenin B is indeed identical with albanin F, our present paper implies revision of the structure assigned to moracenin B.

(Received September 18, 1980)