(-)-Grandisin from Cryptocarya crassinervia

J. M. Saad, E. Soepadamo, X.-P. Fang, J. L. McLaughlin, and P. E. Fanwick

J. Nat. Prod., **1991**, 54 (6), 1681-1683• DOI:
10.1021/np50078a033 • Publication Date (Web): 01 July 2004

Journal of Natural Products
Vol. 54, No. 6, pp. 1681-1683, Nov-Dec 1991

(−)-GRANDISIN FROM *CRYPTOCARYA CRASSINERVIA*

J.M. SAAD, E. SOEPADAMO,

Department of Biochemistry and Department of Botany, University of Malaya, 59100 Kuala Lumpur, Malaysia

X.-P. FANG, J.L. MCLAUGHLIN,*

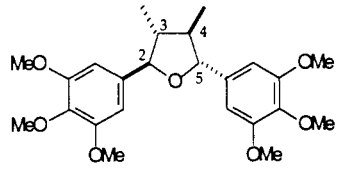
Department of Medicinal Chemistry and Pharmacognosy, School of Pharmacy and Pharmacal Sciences

and P.E. FANWICK

Department of Chemistry, School of Sciences, Purdue University, West Lafayette, Indiana 47907

ABSTRACT.—The known lignan (−)-grandisin [**1**] has been isolated from *Cryptocarya crassinervia* by using the brine shrimp lethality test to direct the isolation; its structure and relative stereochemistry have been determined by ir, ^{1}H nmr, ms, and X-ray crystallography as an all-trans α,α′-diaryl-β,β′-dimethyltetrahydrofuran. Compound **1** is not significantly cytotoxic in our panel of human tumor cells.

In our search for bioactive natural products, a lignan, grandisin [**1**], was isolated from the bark of *Cryptocarya crassinervia* Miq. (Lauraceae). The fractionation was guided by a simple test for lethality to the larvae of brine shrimp (BST) (1). (−)-Grandisin, an α,α′-diaryl-β,β′-dimethyltetrahydrofuran, was first isolated from *Litsea grandis* (Wall) Hook. f. (Lauraceae) (2). Its analogues have been reported as antagonists of platelet-activating factor (PAF), useful for the treatments of inflammation, cardiovascular disorders, asthma, lung edema, adult respiratory distress syndrome, pain, and platelet aggregation (3). We determined the structure and relative configuration of compound **1** by ir, ^{1}H nmr, ms, and X-ray crystallography (Table 2, Figure 1); its X-ray crystallographic structure has not been reported before. This compound showed significant activity in the brine shrimp lethality test, but no significant in vitro activities to human tumor cells were found (Table 1).



1

EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—The mp was uncorrected. The optical rotation was taken with a Perkin-Elmer 241 Polarimeter. The ir spectrum was obtained on a Perkin-Elmer 1600 FTIR spectrometer. ^{1}H-nmr spectra were recorded on a Varian VXR-500S spectrometer. Low resolution ms was obtained on a Finnigan 4000 mass spectrometer, and the hrms was determined on a Kratos MS 50 spectrometer through peak matching.

PLANT MATERIAL.—Bark of *C. crassinervia* was collected from the branches of authenticated trees growing on the peat swamp in Batang Bevjuntai, Selangor, Malaysia. An herbarium sample is preserved at the University of Malaya, Kuala Lumpur, Malaysia.

TABLE 1. Bioactivities of Grandisin [**1**].

BST[a] LC50 (μg/ml)	A-549[b] (ED50 (μg/ml)	MCF-7[c] ED50 (μg/ml)	HT-29[d] ED50 (μg/ml)
21(13–33)	>10	>10	>10

[a]Brine shrimp lethality test with 95% confidence intervals in parentheses.
[b]Human lung carcinoma.
[c]Human breast carcinoma.
[d]Human colon adenocarcinoma.

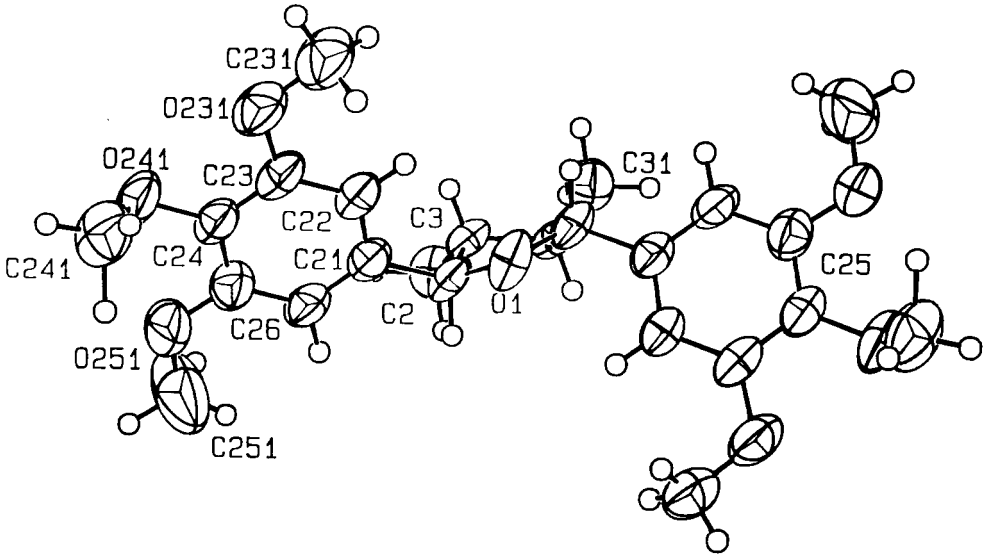


FIGURE 1. ORTEP plot of (−)-grandisin [**1**].

ISOLATION AND IDENTIFICATION.—Dried pulverized bark (600 g) was extracted with 95% EtOH. The extract was rotary evaporated to provide 35 g of brown syrupy extract (F001). F001 was partitioned with $CHCl_3/H_2O$, and the $CHCl_3$ fraction was vacuum-dried to yield 30 g of residue (F003). F003 was further partitioned with 90% MeOH/hexane. The MeOH fraction was then rotary evaporated to yield 26 g of residue (F005). F005 (10 g) was loaded onto a Si gel column and eluted with hexane/$CHCl_3$/MeOH in increasing polarity. The fractions were tested with brine shrimp larvae for bioactivity (1). The active fractions were pooled and rechromatographed on Si gel eluted with hexane/$CHCl_3$/MeOH with increasing polarity; 20-ml fractions were collected and were allowed to evaporate to dryness. Colorless rhombic crystals (ca. 600 mg) were obtained from fractions 15–28. The crystals were washed and recrystallized from hexane/$CHCl_3$.

Grandisin [**1**].—Colorless rhombic crystals: mp 86–88°; $[\alpha]^{25}D$ −46° (c = 0.43, $CHCl_3$); ir (KBr) cm^{-1} 3415, 2955, 1594, 1506, 1459, 1334, 1234, 1127, 1006, 823, 712; cims (NH_3) m/z (%) $[MNH_4]^+$ 450 (6), $[MH]^+$ 433 (59), $[MH-H_2O]^+$ 415 (10), 404 (100); hrcims m/z $[MH]^+$ found 433.2192 for $C_{24}H_{33}O_7$ (calcd 433.2226); eims m/z (%) 368 (7.2), 339 (2.2), 313 (6.3), 299 (3.5), 285 (5.1), 236 (9.4), 179 (6.4), 129 (15), 117 (23), 91 (17), 83 (55), 69 (100); ^1H nmr ($CDCl_3$) δ 6.61 s (H-2′, -6′, -2″, -6″), 4.63 d (H-2, -5, J = 9.4 Hz), 3.87 s (3′-, 5′-,3″-,5″-OMe), 3.82 s (4′-,4″-OMe), 1.77 m (H-3,-4), 1.05 d (3-,4-Me, J = 5.50 Hz).

X-RAY CRYSTALLOGRAPHIC ANALYSIS OF (−)-

TABLE 2. Atomic Fractional Coordinates (ESD) of Grandisin [**1**].[a]

Atom	x	y	z	Atom	x	y	z
O-1	0.8033(2)	1.60660	¼	C-23	0.7352(3)	1.5455(3)	0.0806(2)
O-231	0.6338(2)	1.4961(2)	0.0573(1)	C-24	0.8197(3)	1.5616(3)	0.0455(1)
O-241	0.7984(2)	1.5325(2)	−0.0094(1)	C-25	0.9252(3)	1.6112(3)	0.0658(2)
O-251	1.0031(2)	1.6211(3)	0.0287(1)	C-26	0.9465(3)	1.6432(3)	0.1201(2)
C-2	0.8896(3)	1.6736(3)	0.2131(1)	C-31	1.0222(3)	1.8797(4)	0.2023(2)
C-3	0.9081(3)	1.7883(3)	0.2197(1)	C-231	0.5434(4)	1.4731(5)	0.0918(2)
C-21	0.8633(3)	1.6305(3)	0.1544(1)	C-241	0.7774(4)	1.4250(4)	−0.0203(2)
C-22	0.7582(3)	1.5816(3)	0.1352(1)	C-251	1.1126(5)	1.6720(7)	0.0460(3)

[a]Only half of the carbon and oxygen atoms of the molecule were anisotropically refined because it is a twofold symmetrical molecule; half of the hydrogen atoms were also refined isotropically.

GRANDISIN [**1**].[1]—*Crystal data.*—$C_{26}H_{36}O_{7.5}$, M = 468.57 (the actual molecular formula and mol wt are $C_{24}H_{32}O_6$ and 432.22, but the crystal formula and formular weight for X-ray data collection contained a part of a solvent molecule). Hexagonal, $a = 14.091$ (1), $c = 24.183$ (s) Å, $V = 4158.2$ Å^3 (by least-squares refinement, using the setting angles of 25 reflections in the range $12 < \theta < 15°$, measured by the computer-controlled diagonal slit method of centering), λ (Mo-Kα) = 0.71073 Å, space group *P6(1)22* (No. 178), $Z = 6$, $Dx = 1.10\ g \cdot cm^{-3}$. Crystal dimensions 0.63 × 0.38 × 0.35 mm, μ (Mo-Kα) = 0.75 cm^{-1}.

Data collection and processing.—Enraf-Nonius CAD4 diffractometer, ω/2θ mode with w scan width = 0.68 + 0.350 tan θ, 2θ range 4.00–50.00°, take-off angle 2.95°, ω scan rate 2–20° min^{-1}, graphite-monochromated Mo-Kα radiation; 2832 reflections measured (*h*, *k*, *l* limits: −16–0, 0–14, 0–28), 2462 unique, giving 1095 with $I > 3.0\ \sigma(I)$. Corrections were applied for Lorentz and polarization factors, but not for absorption.

Structure solution and refinement.—The structure was solved using the structure solution program SHELX-86. The remaining atoms were located in succeeding difference Fourier syntheses. Hydrogen atoms were located, and their positions and isotropic thermal parameters were refined. The structure was refined in full-matrix least-squares where the function minimized was $\Sigma w(|Fo| - |Fc|)^2$ and the weight w is defined by the Killean and Lawrence method with terms of 0.020 and 1.0 (4). Scattering factors were taken from Cromer and Weber (5). Anomalous dispersion effects were included in Fc; the values for δf′ and δf″ were those of Cromer and Weber (5). Only the 1095 relections having intensities greater than 3.0 times their standard deviation were used in the refinements. The final cycle of refinement included 211 variable parameters and converged (largest parameter shift was 0.53 times its ESD) with unweighted and weighted agreement factors of: $R1 = \Sigma||Fo| - |Fc||/\Sigma|Fo| = 0.042$, $R2 = SQRT[\Sigma w(Fo - Fc)^2/\Sigma wFo^2] = 0.050$. The standard deviation of an observation of unit weight was 1.14. There were 73 correlation coefficients greater than 0.50. The highest peak in the final difference Fourier had a height of 0.21 e/Å^3 with an estimated error based on δF of 0.04. The refined values for the other enantiomorph are R = 0.042, Rw = 0.050, and the estimated standard deviation of an observation of unit weight is 1.150. Plots of $\Sigma w(|Fo| - |Fc|)^2$ versus $|Fo|$, reflection order in data collection, $\sin\theta/\lambda$, and various classes of indices showed no unusual trends. All calculations were performed on a VAX computer using SDP/VAX.

ACKNOWLEDGMENTS

This work was supported by R01 grant no. CA30909 from the National Cancer Institute, National Institutes of Health, and J.M. Saad acknowledges the support of a visiting Fulbright scholarship.

LITERATURE CITED

1. B.N. Meyer, N.R. Ferrigni, J.E. Putnam, L.B. Jacobsen, D.E. Nichols, and J.L. McLaughlin, *Planta Med.*, **45**, 31 (1982).
2. D. Holloway and F. Scheinmann, *Phytochemistry*, **13**, 1233 (1974).
3. T. Biftu, T.W. Doebber, S.B. Hwang, T.R. Beattie, T.Y. Shen, and R. Stevenson (Merck and Co., Inc.), U.S. Patent No. 4,595,693 (Cl. 514-461; A61K31/34), 17 Jun 1986, Appl. 617,294.04 Jun 1984; *Chem. Abstr.*, **105**, 108508x (1986).
4. R.C.G. Killean and J.L. Lawrence, *Acta Crystallogr., Sect. B*, **25**, 1750 (1969).
5. D.T. Cromer and J.T. Weber, "International Tables for X-ray Crystallography," Kynoch Press, Birmingham, 1974, Vol. 4, Tables 2.2B, 2.3.

Received 23 May 1991

[1]Atomic coordinates for this structure have been deposited with the Cambridge Crystallographic Data Centre and can be obtained on request from Dr. Olga Kennard, University Chemical Laboratory, Lensfield Road, Cambridge CB2 1EW, UK.