



Vaticanol D, a novel resveratrol hexamer isolated from *Vatica rassak*

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Received 13 July 2000; revised 7 August 2000; accepted 10 August 2000

Abstract

Vaticanol D isolated from the bark of *Vatica rassak* is the first instance of a resveratrol hexamer. The structure and relative configuration were established by means of 2D NMR spectroscopy. Vaticanol D possessed a scavenging activity of super oxide. © 2000 Elsevier Science Ltd. All rights reserved.

Keywords: natural product; polycyclic aromatic compound; hexamer; scavenging activity of super oxide.

A simple skeleton of stilbene (C₆–C₂–C₆) adds the occurring numbers in nature (Bryophyta, Pteridophyta and Spermatophyta) owing to the oxidative variation of nucleus and the homogeneous oligomerization ranging from dimer to pentamer.^{1–3} The biological activities of stilbenoids, such as chemopreventive activity of cancer, anti-fungal activity, cytotoxicity against human tumor cell lines, have been revealed by degrees.^{4–6} Stilbenoids are, therefore, regarded as one of important lead compounds for drug development. To find much highly condensed stilbenoid oligomers in nature and to estimate their biological activity is a substantial clue to clarifying their potency. Among the plant kingdom, it is well known that Dipterocarpaceous plants are the abundant resource of stilbenoids.¹ We have already reported the isolation and structure elucidation of six stilbenoids, including three new compounds named vaticanols A–C, from the bark of *Vatica rassak* (Dipterocarpaceae).⁷ Further investigation of the bark resulted in the first reported isolation of a resveratrol hexamer (**1**), vaticanol D.

An acetone extract (60 g) of the dried and pulverized bark (900 g) of *V. rassak* was subjected to column chromatography on silica gel (CHCl₃–MeOH gradient system) to divide the sample into 18 fractions. Further purification of the CHCl₃–MeOH (5:1) fraction by vacuum liquid

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chromatography (CHCl_3 -MeOH=5:1, gradient) and successive preparative TLC (solvent: EtOAc- CHCl_3 -MeOH=60:30:7) achieved the isolation of **1** (8 mg) along with vaticanols A-C.

Vaticanol D (**1**), $[\alpha]_D^{25} +62^\circ$ (MeOH) obtained as a brown solid, showed an $[\text{M}-\text{H}]^-$ ion at m/z 1359 in the negative ion FABMS attributable to the empirical formula $\text{C}_{84}\text{H}_{64}\text{O}_{18}$, and is regarded as a stilbene hexamer.

In the UV spectrum, absorption bands were observed at 228 and 282 nm. The ^1H NMR and 2D-HOHAHA spectral data indicated that the presence of six 4-hydroxyphenyl groups (designated as aromatics A_1 - F_1), a 3,5-dihydroxyphenyl group (A_2), a 3,5-dioxygenated-1,4-disubstituted benzene ring (D_2), two 3,5-dioxygenated-1,2-disubstituted benzene rings (C_2 and F_2), two 3,5-dioxygenated-1,2,6-trisubstituted benzene ring (B_2 and E_2) and four sequences of aliphatic protons in this order (H-7b/H-8b/H-7a/H-8a, H-7c/H-8c, H-7e/H-8e/H-7d/H-8d and H-7f/H-8f) represented by bold lines in Fig. 1. Among them, protons based on a 3,5-dioxygenated-1,4-disubstituted benzene ring (D_2) were observed in an asymmetrical form. The HMQC spectrum was able to complete the assignment of all protonated carbons as shown in Table 1.

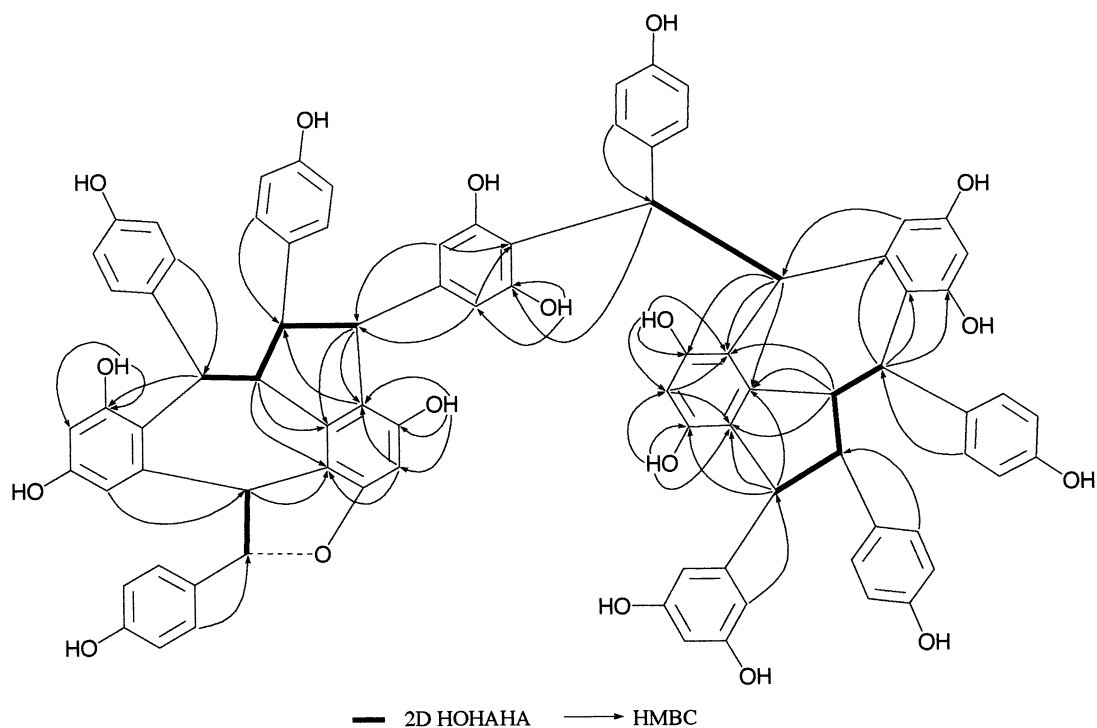


Figure 1. Selected 2D NMR data of **1**

In the HMBC spectrum (Fig. 1), significant correlations via 3J were observed between H-7a/C-2a (6a), H-8a/C-10a (14a), H-7b/C-2b (6b), H-7c/C-2c (6c), H-14c/C-8c, H-7d/C-2d (6d), H-8d/C-10d, H-8d/C-14d, H-7e/C-2e (6e), H-7f/C-2f (6f) and H-14f/C-8f, indicating that the rings A_1 , A_2 , B_1 , C_1 , C_2 , D_1 , D_2 , E_1 , F_1 and F_2 were attached to C-7a, C-8a, C-7b, C-7c, C-8c, C-7d, C-8d, C-7e, C-7f and C-8f, respectively. Considering the correlations between H-7c/C-11d and H-7e/C-11f, the ring D_2 and F_2 could be connected with C-7c and C-7e, respectively. The remaining two benzene rings (B_2 and E_2) were attached as shown in Fig. 1 by a detailed analysis of the HMBC spectrum. Although a long-range correlation between

Table 1
 ^1H and ^{13}C NMR^a spectral data of vaticanol D (**1**)

No.	δ_{H}	δ_{C}	No.	δ_{H}	δ_{C}
1a		133.7	1d		130.2 ^e
2a, 6a	6.91 (d, 8.8)	131.1	2d, 6d	7.10 (d, 8.8)	131.3
3a, 5a	6.56 (d, 8.8)	115.4 ^d	3d, 5d	6.76 (d, 8.8)	115.4 ^d
4a (OH)	7.97 (br s)	156.5	4d (OH)	8.17 (br s)	156.3 ^c
7a	3.79 (t, 7.0)	61.8	7d	4.00 (dd, 10.3, 9.3)	57.3
8a	4.44 (d, 7.0)	54.9	8d	3.50 (d, 10.3)	50.6
9a		148.2	9d		143.2
10a, 14a	6.13 (d, 2.0)	107.1	10d	6.22 (d, 2.0)	108.7
11a, 13a (OH)	7.94 (br s)	159.1	11d (OH)	8.02 (br s)	157.87
12a	6.13 (t, 2.0)	101.4	12d		119.2
			13d (OH)	n.o.	157.1
			14d	6.33 (d, 2.0)	108.5
1b		139.6	1e		132.8
2b, 6b	6.93 (d, 8.8)	130.2 ^e	2e, 6e	5.97 (d, 8.5)	132.9
3b, 5b	6.42 (d, 8.8)	114.74 ^b	3e, 5e	6.27 (d, 8.5)	113.7
4b (OH)	7.72 (br s)	155.0	4e (OH)	7.83 (br s)	155.9
7b	4.68 (d, 7.0)	39.8	7e	4.78 (br s)	42.4
8b	4.79 (t, 7.0)	54.4	8e	4.23 (dd, 9.3, 3.0)	51.1
9b		148.8	9e		143.8
10b		122.9	10e		116.1
11b (OH)	7.55 (br s)	154.7	11e		160.4
12b	5.87 (s)	103.7	12e	6.01 (s)	96.3
13b (OH)	6.38 (br s)	152.1	13e (OH)	5.92 (br s)	155.4 ^f
14b		122.2	14e		123.6
1c		137.3	1f		133.6
2c, 6c	7.37 (d, 8.3)	131.4	2f, 6f	7.52 (d, 8.8)	130.6
3c, 5c	6.42 (d, 8.3)	114.69 ^b	3f, 5f	6.97 (d, 8.8)	116.5
4c (OH)	7.70 (br s)	155.4 ^f	4f (OH)	8.62 (br s)	158.6
7c	6.02 (d, 12.8)	40.7	7f	5.63 (d, 9.3)	95.5
8c	6.24 (d, 12.8)	45.8	8f	5.29 (d, 9.3)	52.4
9c		147.0	9f		140.3
10c		120.7	10f		123.5
11c (OH)	7.31 (br s)	156.7	11f (OH)	7.88 (br s)	157.90
12c	5.89 (d, 2.0)	100.5	12f	6.12 (d, 2.0)	102.0
13c (OH)	7.59 (br s)	155.8	13f (OH)	7.96 (br s)	156.4 ^c
14c	6.62 (d, 2.0)	105.11	14f	6.09 (d, 2.0)	107.9

^a ^1H and ^{13}C NMR spectra were measured in acetone- d_6 at 500 and 125 MHz, respectively. All protons and carbons were assigned by the HMQC and HMBC spectra. ^{b,c} Interchangeable; ^{d-f} overlapping; n.o., signal not observed.

H-7f/C-11e was not observed, it could be confirmed that a dihydrofuran ring existed in **1** by considering the molecular formula. Finally the planar structure of vaticanol D was drawn as shown in Fig. 1. The relative stereochemistry was determined by a ROESY spectrum as shown in Fig. 2. The protons H-7c and H-8c were in a *trans* conformation since $J_{7c,8c} = 12.8$ Hz. The occurrence of a number of stilbene tetramers and one instance of a pentamer in nature has been reported (e.g. *Carex* (Cyperaceae),^{8,9} *Vitis* (Vitaceae)¹⁰ and *Sophora* (Leguminosae)³), but the hexamer mentioned above has been first isolated from plants.

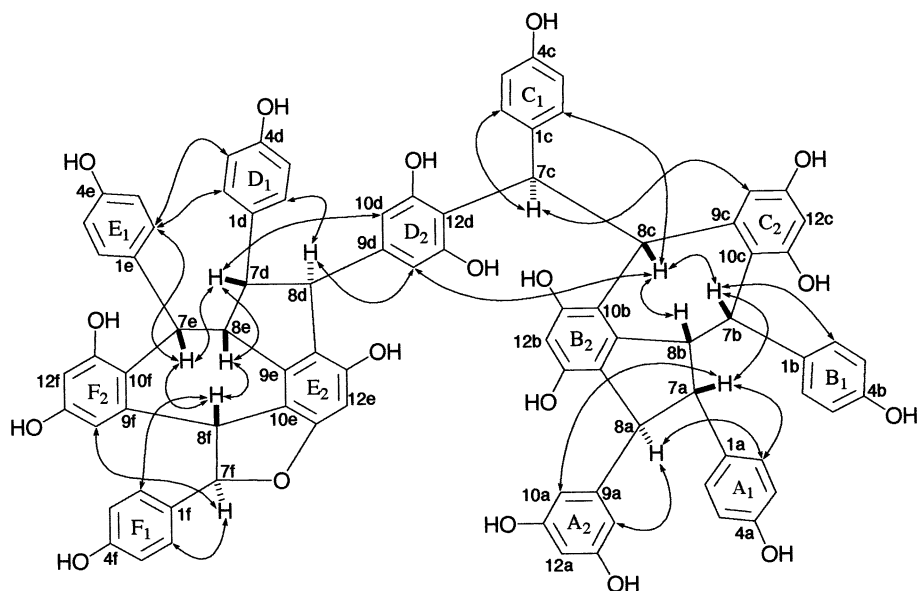


Figure 2. ROESY correlations in **1**

An acetone extract of *V. rassak* showed a scavenging activity for super oxide (88% at 1% concentration of extract) in a xanthin–xanthin oxidase system.^{11,12} Among the isolates (vaticanols A–D), vaticanol D showed a scavenging activity of super oxide at IC₅₀ 7.4 μM.

Acknowledgements

The authors (M.I. and K.T.) thank a Grant-in-Aid for Scientific Research on Priority Area (08281105) to support this work in part.

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