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STRUCTURE OF RISHITINONE, A STRESS METABOLITE FROM DISEASED POTATO TUBERS[1)]

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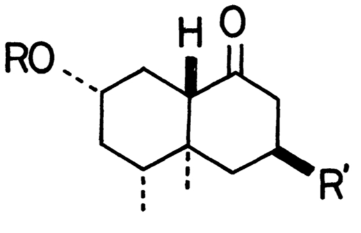
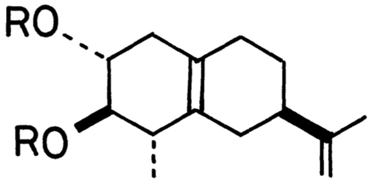
Structure and configuration of rishitinone, a stress metabolite isolated from diseased potato tubers, was determined to be formula 1 on the basis of the chemical and spectral evidence.

In a continuing study on phytoalexins produced by tuber tissues of potatoes (*Solanum tuberosum* × *S. demissum*) infected by an incompatible race of *Phytophthora infestans*,2) we isolated a new sesquiterpene, designated as rishitinone (1), from the chloroform extracts of the tissues, which was not detected in the corresponding extracts of healthy tissues and showed a comparable fungitoxicity with that of rishitin2) (2). We describe herein the isolation and structure elucidation of the stress metabolite (1).

Neutral chloroform extracts (80 g), obtained from the diseased potato tuber tissues (300 kg), were separated as described previously2) to give "rishitin-rich fraction" (Fraction F,2) 5.5 g), which on acetylation followed by chromatogarphy afforded a semi-crystalline mixture of acetates. Fractional recrystallization of the mixture gave pure rishitin diacetate2) (2a, 3.3 g), mp 65-67 °C, and crude oily rishitinone acetate (1a, 0.11 g), which was submitted to purification by chromatography to yield rishitinone acetate (1a), oily and $[\alpha]_D$ -2.4°,3) in pure state. The acetate (1a) was then hydrolyzed (K_2CO_3 in CH_3OH, room temp, 1 h) to give rishitinone (1), mp 72-75 °C and $[\alpha]_D$ +10.1°.



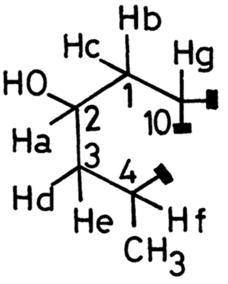
1 R=H, R'=$C(CH_3)=CH_2$
1a R=Ac, R'=$C(CH_3)=CH_2$
3 R=H, R'=$CH(CH_3)_2$



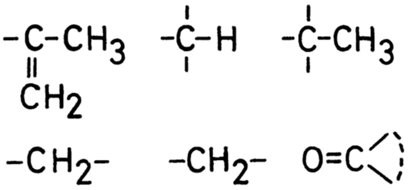
2 R=H
2a R=Ac

Rishitinone (1) was analyzed for $C_{15}H_{24}O_2$ (m/e 236, M^+) and converted by hydrogenation (Pd-C in C_2H_5OH) into dihydrorishitinone (3), mp 80-82 °C and $[\alpha]_D$ +7.2°. The IR and ^{1}H-NMR spectra3) indicated the presence of the following structural units: a *t*-methyl group [1, δ 0.76 (3H, s)]; a *s*-methyl [1, δ 0.84 (3H, d, J = 7 Hz)]: an isopropenyl [1, ν_{max} 1615 and 887 cm^{-1}; δ 1.76 (3H, s) and 4.77 (2H, br s); 3, ν_{max} 1388 and 1372 cm^{-1}, δ 0.91 (6H, d, J = 6 Hz)], a carbonyl [1, ν_{max} 1710 cm^{-1}]; a *sec*-hydroxyl [1, ν_{max} 3500 cm^{-1}; δ 3.65 (1H, m, W_H = 25 Hz); 1a, ν_{max} 1739 and 1248 cm^{-1}; δ 2.02 (3H, s) and 4.77 (1H, m, W_H = 25 Hz)]. The ^{13}C-NMR spectra obtained under proton-noise decoupled and single-frequency off-resonance decoupled conditions [δ 12.9, 15.5, and 20.4 (each $-CH_3$); 30.2, 39.3, 42.8, and 46.0 (each $-CH_2-$); 41.3, 41.5, 56.1, and 69.5 (each $-\overset{|}{C}H-$); 40.7 (■)3); 109.9 ($=CH_2$); 147.4 ($=\overset{|}{C}-$); 211.1 ($-\overset{|}{C}=O$)] provided additional information of carbon atoms, indicating that the ketone (1) possesses a bicyclic skeleton.

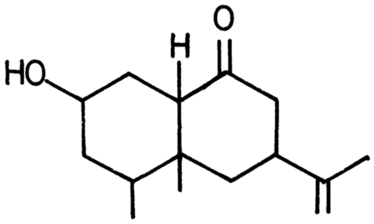
The ^1H-NMR spectra of 1 in the presence of the shift reagent $Eu(dpm)_3$ (0.2 and 0.4 mol equiv) effected downfield shifts of all the signals, and exhibited one-proton broad signals (W_H = 25 Hz) at δ 6.80 and 10.50 in the respective spectra, which were reasonably assigned to the proton on the carbon bearing the hydroxyl group. These spectra, coupled with spin-decoupling studies (Table 1), strongly suggested that the ketone (1) would probably involve a partial formula A. In view of the remaining nine carbon units (formula B) deduced from the afore-mentioned



A



B



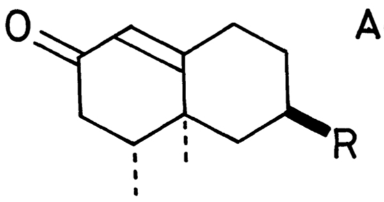
C

Table 1. The ^1H-NMR spectra of rishitinone (1) in the presence of the shift reagent $Eu(dpm)_3$ (CCl_4, 100 MHz) and results of the spin-decoupling studies

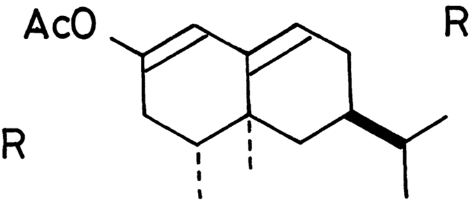
Protons irradiated (δ and Hz)	Observed multiplicity change (δ and Hz)
Mole ratio of 1:$Eu(dpm)_3$ 5:1	
6.80 [Ha (C-2), br (W_H = 25)]	4.00 [Hb, dt → dd (J = 12 and 4)]
	3.20 ∿ 3.80 [Hc ∿ He, br m → changed]
4.00 [Hb (C-1), dt (J = 12 and 4)]	6.80 [Ha, br → changed]
3.50 [Hc ∿ He (C-1 ∿ C-3), br m]	6.80 [Ha, br → changed]
	2.30 [Hf, br → changed]
2.34 [Hf (C-4), br]	1.23 [CH_3, d → s]
1.23 [CH_3 (C-4), d (J = 7)]	2.30 [Hf, br → dd (J = 11 and 3)]
Mole ratio of 1:$Eu(dpm)_3$ 5:2	
10.5 [Ha (C-2), br (W_H = 25)]	5.6 ∿ 6.4 [Hb ∿ He, br m → changed]
6.12 [Hb and Hc (C-1), br m]	4.12 [Hg, dd → s]
4.12 [Hg (C-10), dd (J = 12 and 4)]	6.0 ∿ 6.3 [Hb ∿ Hc, br m → changed]

spectra, specially from the ^{13}C-NMR spectra, formula C is presented as one of the most plausible planar structures for **1**. This structure was confirmed by correlation between reshitinone (**1**) and (+)-nootkatone$^{4)}$ (**4**).

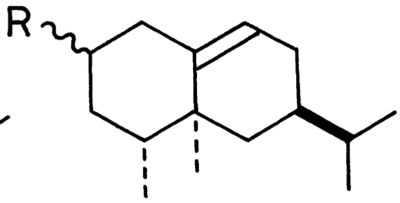
(+)-Nootkatone (**4**) was submitted to partial hydrogenation in benzene containing tris(triphenylphosphine)chlororhodium (room temp, 14 h) to give dihydronootkatone (**5**), oily and $[\alpha]_D$ +167°, in an 81% yield; ν_{max} 1670 cm^{-1}; δ 0.69, 0.72, and 0.78 (each 3H, d, J = 7 Hz, 12-, 13-, and 14-H), 1.08 (3H, s, 15-H), and 5.78 (1H, s, 1-H). The compound (**5**) was converted by a modification of the Edward procedure$^{5)}$ into dienol acetate (**6**), oily and $[\alpha]_D$ +45.3°, in a 69% yield; m/e 262 (M^+); ν_{max} 1759 and 1664 cm^{-1}; δ 2.12 (3H, s, $OCOCH_3$), 5.42 (1H, m, W_H = 10 Hz, 9-H), and 5.71 (1H, s, 1-H), which on hydride reduction ($NaBH_4$) produced a 1:5 mixture of homoallylic alcohols (**7**) and (**8**). The major product (**8**) was assigned an equatorial alcohol on the basis of the following spectral data: **7**, oily and $[\alpha]_D$ -30.7°; m/e 222 (M^+); ν_{max} 3300, 1382, and 1365 cm^{-1}; δ 3.99 (1H, m, W_H = 8 Hz, 2-H) and 5.43 (1H, m, W_H = 12 Hz, 9-H): **8**, oily and $[\alpha]_D$ -34.2°; m/e 222 (M^+); ν_{max} 3320, 1381, and 1368 cm^{-1}; δ 3.56 (1H, m, W_H = 25 Hz, 2-H) and 5.38 (1H, m, W_H = 12 Hz, 9-H). The hydroxyl group of the latter (**8**) was then protected with <u>t</u>-butyldimethylsilyl (TBDMS) chloride and imidazole in dimethylformamide. The resulting silyl ether (**8a**), oily and $[\alpha]_D$ -16.3°; m/e 337 (M^+ + 1); ν_{max} 1100 and 1258 cm^{-1}, obtained in a 76% yield, underwent hydroboration followed by oxidation with alkaline hydrogen peroxide, producing a mixture of two isomeric alcohols (**9**) and (**10**) and the starting alkene (**8a**), which were separated by chromatography over silica gel in 23 (crude), 42 (pure), and 8% yields, respectively. The major product (**10**) was assigned 9β-hydroxy-<u>trans</u>-valencane structure on the basis of the spectral data and the steric manner (<u>cis</u>-addition from the less-hindered side) of this reaction:$^{6)}$ **10**, oily and $[\alpha]_D$ +38.0°; m/e 354 (M^+); ν_{max} 3360, 1258, and 1100 cm^{-1}; δ 3.49 (1H, dt, J = 4.5 and 10 Hz, 9-axial-H) and 3.58 (1H, m, W_H = 25 Hz, 2-H); acetate (**10a**) of **10**, oily and $[\alpha]_D$ +35.5°; δ 3.76 (1H, m, W_H = 25 Hz, 2-H) and 4.74 (1H, dt, J = 4 and 11 Hz, 9-H). The <u>trans</u>-decalol (**10**) was then oxidized with Sarett reagent (room temp, 15 min) to yield the corresponding ketone (**11**) in a quantitative yield, mp 51-53 °C and $[\alpha]_D$ +2.1°; m/e 352 (M^+); ν_{max} 1717 cm^{-1}; δ 0.66 (3H, s, 15-H) and 0.90 (9H, br m, 12-, 13-, and 14-H), which was hydrolyzed without epimerization at C-10 in a 3:1:1 mixture of acetic acid, water, and tetrahydrofuran to give dihydrorishitinone (**3**) in a 95% yield, identical with an authentic specimen derived from natural (+)-rishitinone in all respects. Supporting the assigned configuration, the ORD spectrum$^{3)}$ of the ketone (**3**) showed a typical positive Cotton effect, whose amplitude (a = +26.6°) was in good accord with that (a = +26°) reported for a known ketone$^{7)}$ (**12**), which had been derived from natural (-)-aristolochene, one of valencenes. All these results indicate the structure and configuration of (+)-rishitinone to be represented correctly by formula **1**.



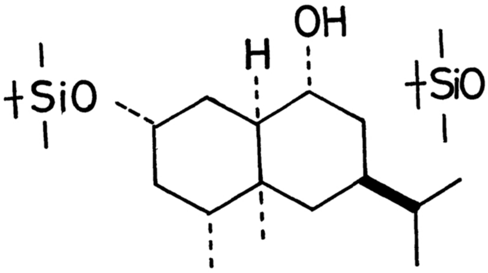
4 $R=C(CH_3)=CH_2$
5 $R=CH(CH_3)_2$



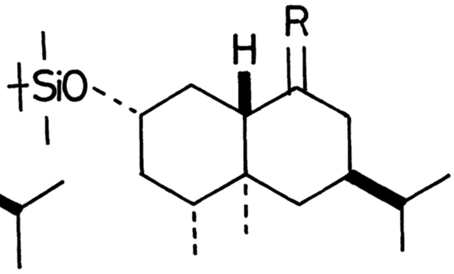
6



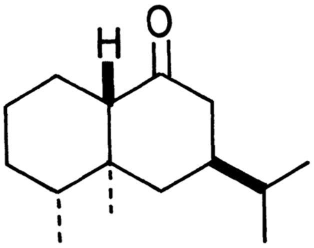
7 R=β-OH
8 R=α-OH
8a R=α-OTBDMS



9



10 R=β-OH, α-H
11 R=O



12

References and Notes

1) Part XXV of "Studies on the Phytoalexins;" Part XXIV, A. Murai, H. Taketsuru, and T. Masamune, Bull. Chem. Soc. Jpn., 53, 1049 (1980).
2) T. Masamune, A. Murai, M. Takasugi, A. Matsunaga, N. Katsui, N. Sato, and K. Tomiyama, Bull. Chem. Soc. Jpn., 50, 1201 (1977).
3) a) The optical rotations were obtained in ethanol. The IR and NMR spectra were measured in liquid state (oil) or in Nujol (solid), and in chloroform-d, respectively, unless otherwise stated. b) The abbreviation ■ denotes a carbon atom bearing no hydrogen atom(s).
4) W. D. MacLeod, Jr., Tetrahedron Lett., 1965, 4779.
5) B. E. Edwards and P. N. Rao, J. Org. Chem., 31, 324 (1966).
6) H. C. Brown and G. Zweifel, J. Am. Chem. Soc., 83, 2544 (1961); D. J. Pasto and F. M. Klein, J. Org. Chem., 33, 1468 (1968).
7) T. R. Govindachari, P. A. Mohamed, and P. C. Parthasarathy, Tetrahedron, 26, 615 (1970).

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