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Lingans from Korean Red Ginseng

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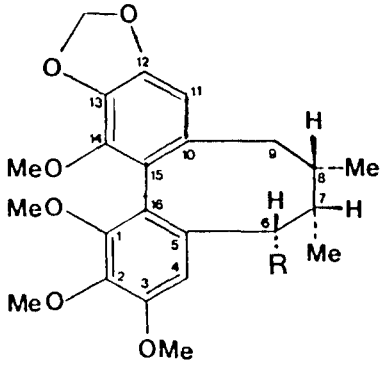
Abstract□Two lingans were isolated from hexane-soluble fraction of Korean red ginseng. Their chemical structures were elucidated as gomisin N and gomisin A by spectrometric analysis.

Keywords□*Panax ginseng* C.A. Meyer, Araliaceae, lignan, dibenzocyclooctane, gomisin N gomisin A.

We reported the isolation of some phenolic substances such as maltol, salicylic acid, vanillic acid and *p*-coumaric acid from the extract of *Panax ginseng* C.A. Meyer as the effective components for the antioxidant and antifatigue activities[1,2]. Others described detection of caffeic acid[3], and isolation of ferulic acid[4] as the antioxidant substances.

Besides the phenolic substances, some papers described the isolation of some polyacetylenes as the cytotoxic substances from the hexane soluble fraction of ginseng extract[5-9]. Our continuing effort to isolate new phenolic substances from the hexane soluble fraction of Korean ginseng led us to isolate compound I, $C_{23}H_{28}O_6$ mp. 108-110°C and compound II, $C_{23}H_{28}O_7$ mp 50-52°C. Present paper is concerned with the identification of the two compounds as the known lignans, gomisin-N[10,11] and gomisin-A[12,13] isolated from *Schizandra chinensis,* having antihepatotoxic activities[14,15].



R:H gomisin N
R:OH gomisin A

EXPERIMENTAL

Melting points were determined on Mitamura-Riken apparatus and are uncorrected. UV and IR spectra were recorded by Gilford System 2600 UV-VIS spectrophotometer, and by Perkin-Elmer 283B, respectively. NMR spectra were taken at using TMS as an internal standard on Bruker 300, JEOL 270, JEOL 500 and Varian FT 80A. Mass spectra were measured with Hewlett Pachard Model HP 5985B.

Following chromatographic solvents were used. Solvent: A (Hex./EtOAc = 20 : 1), B (Hex./EtOAc = 10 : 1), C (Hex./EtOAc = 6 : 1), D (Benz./EtOH = 20 : 1), E (Benz./EtOH = 85:15)

Extraction and fractionation

Powdered Korean red ginseng (5.3 kg) was extracted with 5 *l* of hexane for three times to give 59g of hexane extract. Fig. 1 shows the TLC pattern of the hexane extract visualized by the spray of 10% H_2SO_4 and heating. The hexane extract (13g) was chromatographed on silica gel column (Art. 7734; 400g) eluting by solvent A and divided into five fractions, fraction 1-5, depending on its TLC pattern.

Isolation of compound I

Fr. 2 (750 mg) was chromatographed on silica gel column (Art. 7734, 20g) with solvent B to give Fr. 2-B (180 mg), which was chromatographed again on silica gel column (Art. 7729, 20g) using solvent D to give Fr. 2-B-b, from which compound I was crystallized as prizm. mp, 108-110°C; Mass [EI] (rel. Int. %): *m/z* 400 (100) [M+], 385 ($M^+ - CH_3$); UV (EtOH) λ_{max} nm (log ε): 252 (3.22), 285 (2.72); IR (cm^{-1}, KBr): 1500-1600 (aromatic); ^{1}H-NMR (300 MHz, $CDCl_3$): shown in Table I; ^{13}C-NMR (75

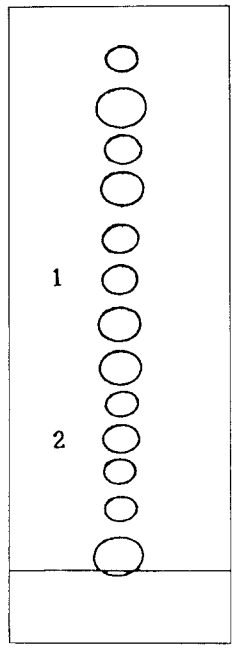


Fig. 1. TLC pattern of hexane extract of Korean red ginseng. Plate, precoated silica gel (E. Merk, type F 254); Developing solvent, hexane/ethylacetate (4:1); coloring agent, 10% H_2SO_4; 1, compound I; 2, compound II.

MHz, $CDCl_3$): shown in Table II.

Isolation of compound II

The Fr. 4 (700 mg containing black spot on Rf. 0.3) was concentrated and chromatographed on silica gel column (Art. 7734, 70g) with solvent C to give Fr. 4-B (160 mg), which was rechromatographed on silica gel column (Art. 9385, 20g) eluting by solvent E to give Fr. 4-B-b. Compound II was obtained as colorless needles. (yield 32 mg). From Fr. 4-B-b, mp. 50-52°C; Mass [EI] (rel. Int. %): *m/z* [M+] 416 (100), 345 (11.1), 314 (28.9), 243 (15.5); UV (EtOH) λ_{max} (log ε): 253 (3.68), 2.79 (3.16); IR (cm^{-1}, KBr): 3500, 1500-1600; ^{1}H-NMR ($CDCl_3$, 270 MHz): shown in Table III; ^{13}C-NMR ($CDCl_3$, 67.5 MHz): shown in Table IV.

RESULTS AND DISCUSSION

Identification of compound I

Compound I, [M+] *m/z* 400 was shown to be aromatic compound, since it shows UV absorption maxima on 252 and 285 nm and aromatic absorption peaks of 1500-1600 cm^{-1} on its IR-spectrum. ^{1}H-NMR spectrum of compound I shows 28 protons which were assigned as twelve protons of four methoxy groups,

Table I. ^{1}H-NMR of compound I from *Panax ginseng* and gomisin N

No. of proton	Comp. I (300 MHz)	Gomisin N[10] (100 MHz)
4-H	6.55 (1H, s)	6.55 (1H, s)
6-H_{AB}		2.57 (2H, m)
6-H_A, 9-H_B	2.56 (2H, m)	
7-H, 8-H	1.83 (2H, m)	1.83 (2H, m)
6-H_B	2.03 (1H, dd, J = 13.5 & 1)	
9-H_A	2.27 (1H, dd, J = 13.5 & 8)	2.27 (1H, dd, J = 13.5 & 8)
9-H_B		2.03 (1H, dd, J = 13.5 & 1)
11-H	6.48 (1H, s)	6.47 (1H, s)
17, 18-CH_3	0.73 (3H, d, J = 7)	0.73 (3H, d, J = 7)
	0.73 (3H, d, J = 7)	0.73 (3H, d, J = 7)
	0.97 (3H, d, J = 7)	0.97 (3H, d, J = 7)
CH_2 (O-, O-)	5.93 (2H, s)	5.93 (2H, s)
OCH_3	3.55 (3H, s)	3.55 (3H, s)
	3.83 (3H, s)	3.82 (3H, s)
	3.93 (6H, s)	3.93 (6H, s)

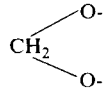
two protons of one methylene-dioxyl group, six protons of two aliphatic methyl groups, two singlet aromatic protons and six overlapped protons of aliphatic methine and methylene nature assigned by DEPT spectrum.

^{13}C-NMR spectrum of compound I shows 23 carbons which were assigned as twelve aromatic carbons (lower field than δ 102.9), two methine carbons at δ 40.7 and 33.7, two methylene carbons at δ 35.4 and 39.0 and two methyl carbons at δ 21.5 and 12.7 together with one methylene-dioxy carbon at ~100.7 and four methoxy carbons at δ 61.0, 60.5, 59.6 and 55.8.

Based on the above spectral data, the chemical structure of compound I was roughly envisaged as being a substituted lignan compound which has four mothoxyl groups and one methylene-dioxy group on its aromatic rings of lignan skeleton composed of two units of C_6-C_3 (phenylpropanoid).

When we consider that the total number of aromatic substitution is observed as eight (four methoxyl, one methylene-dioxyl and two aromatic protons) from the PMR spectra and the unsaturation degree

Table II. ^{13}C-NMR data of compound I from *Panax ginseng* and gomisin N

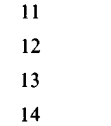
No. of Carbon	Comp. I (75 MHz)	Gomisin N[11] (15.04 MHz)
1	151.5	151.7
2	139.9	140.2
3	151.5	151.6
4	110.7	110.7
5	134.0	134.1
6	39.0	39.2
7	33.7	33.6
8	40.7	40.8
9	35.4	35.6
10	137.8	137.8
11	102.9	102.9
12	148.6	148.7
13	134.5	134.6
14	141.0	141.1
15	121.2	121.4
16	123.2	123.4
17	21.5	21.5
18	12.7	12.9
	100.7	100.7
C_2-OCH_3	61.0	61.0
C_1-OCH_3	60.5	60.5
C_{14}-OCH	59.6	59.6
C_3-OCH_3	55.8	55.9

of compound I, we could obtain further structural informations that the two benzene rings are participating in the cyclooctane ring produced by fusion of two C_6-C_3 units. By the reference studies, we found that the lignans from *Schizandra chinensis* are falling into afore said lignan skeleton and finally we found that ^{1}H-NMR and ^{13}C-NMR data of compound I were identical with those of gomisin-N found in reference (Tables I and II).

Identification of compound II

The spectral data of compound II are very similar to those of compound I except that compound II has one additional oxygen atom than compound I by showing 16 mass unit higher. The additional oxygen atom was identified as hydroxyl group based on its

Table III. ^{1}H-NMR data of compound II from *Panax ginseng* and gomisin A

No. of proton	Comp. II (270 MHz)	Gomisin N[12, 13] (60 MHz)
4-H_A	2.37 (1H, d, J = 13.5)	2.50 (2H, ABq, J_{AB} = 13.5)
6-H_B	2.69 (1H, d, J = 13.5)	
7-OH	1.87 (1H, s)	1.85 (1H, s)
8-H	1.89 (1H, m)	1.80 (1H, m)
9-H_A	2.34 (1H, d, J = 14.0)	2.44 (2H, ABX_{octet},
9-H_B	2.59 (1H, d, J = 14.0)	J_{AB} = 14, J_{AX} = 2, J_{BX} = 6)
17-CH_3	1.25 (3H, s)	0.80 (3H, d, J = 7)
18-CH_3	0.83 (3H, d, J = 7.2)	1.25 (3H, s)
	5.96 (2H, s)	5.96 (2H, s)
$-OCH_3$	3.50 (3H, s)	3.50 (3H, s)
	3.80 (3H, s)	3.80 (3H, s)
	3.90 (6H, s)	3.93 (6H, s)
aromatic	6.48 (1H, s)	6.49 (1H, s)
proton	6.62 (1H, s)	6.62 (1H, s)

IR absorption at 3500 cm^{-1} and deutrated ^{1}H-NMR spectra of compound II. Based on the same logical treatment as compound I, compound II was identified as gomisin-A which has already been described as antihepatotoxic compound of *Schizandra chinensis*. The ^{1}H-NMR and ^{13}C-NMR data of compound II are tabulated in Table III and IV respectively.

From the hexane soluble fraction of *Panax ginseng*, in conclusion, we isolated compound I, $C_{23}H_{28}O_6$, mp 108-110°C and compound II, $C_{23}H_{28}O_7$, mp 50-52°C by repeated column chromatographic purification. By spectrometric analysis, they were identified as gomisin N and gomisin A, which have been described already as antihepatotoxic components from *Schizandra chinensis* Baill.

ACKNOWLEDGEMENT

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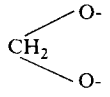
Table IV. ^{13}C-NMR data of compound II from *Panax ginseng* and gomisin A

No. of Carbon	Comp. II (67.5 MHz)	Gomisin A[12] (20 MHz)
1	152.1	152.1
2	140.8	140.8
3	152.3	152.3
4	110.4	110.4
5	132.1	132.1
6	40.6	40.6
7	71.7	71.1
8	42.1	42.1
9	33.8	33.8
10	132.5	132.5
11	105.9	105.9
12	147.9	147.9
13	135.0	135.0
14	141.3	141.3
15	121.9	121.9
16	124.2	124.2
17	30.1	15.8
18	15.8	30.1
	100.8	100.8
C_2-OCH_3	61.0	61.0
C_1-OCH_3	60.6	60.6
C_{14}-OCH_3	59.6	59.6
C_3-OCH_3	55.8	55.0

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