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Studies on the Agarwood (Jinkō). I. Structures of 2-(2-Phenylethyl) chromone Derivatives[1)]

YASUO SHIMADA,[a] TAKAE TOMINAGA,[b] TENJI KONISHI[b] and SHIU KIYOSAWA*,[b]

Mitsuboshi Seiyaku Company,[a] 153, Gose-shi, Nara, 639-22, Japan and Kyoto College of Pharmacy,[b] Nakauchi-cho, Misasagi, Yamashina-ku, Kyoto 607, Japan

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Six kinds of chromone derivatives, named AH_1, AH_2, AH_3, AH_4, AH_5 and AH_6, were isolated from agarwood (Jinkō) from Kalimantan. Four constituents, AH_3, AH_4, AH_6 and AH_5, were characterized as 6-hydroxy- (I), 6-methoxy- (II) and 6,7-dimethoxy-2-(2-phenylethyl)chromone (IV) and 6-methoxy-2-[2-(3-methoxyphenyl)ethyl]chromone (III), respectively.

Keywords——2-(2-phenylethyl)chromone derivatives; agarwood, Aquilariaceae; ^{13}C-NMR spectrum

Agarwood (Jinkō) has been used as an incense from ancient times in the Orient. The structures of several of its constituents, several sesquiterpenes, an oxygenated chromone derivative and others, have been reported.[2—4)]

Many kinds of agarwood have appeared on the market, but it is very difficult to find a basis for evaluating their quality. In the course of a study to evaluate the quality of agarwood products, six substances named AH_1, AH_2, AH_3, AH_4, AH_5 and AH_6 were isolated from the ether and acetone extracts of agarwood from Kalimantan. In this paper, the isolation of AH_1—AH_6 and the structures of AH_3—AH_6 are described. The isolation procedure is shown in Chart 1. Fluorescence under ultraviolet light is very effective for detecting these compounds on thin-layer chromatograms (TLC).

AH_3(I), $C_{17}H_{14}O_3$, mp 224—225°C, was suggested to be a chromone derivative by its infrared (IR) spectrum, and it exhibited the signals of one hydroxyl and one phenethyl group in the proton nuclear magnetic resonance (^{1}H-NMR) spectrum, as shown in Table I. The ultraviolet (UV) spectrum indicated that the hydroxyl and phenethyl groups should be located at C_6 and C_2 of the chromone owing to the distinct absorption maxima at 226, 240 and 332 nm.[5)] These conclusions were confirmed by the ^{13}C-NMR spectrum, in which the signals of C_2 (168.32 ppm) and C_6 (156.24 ppm) appeared at 22.8 and 30.4 ppm downfield from those in chromone, respectively.[6)] As shown in Table II, the assignments of carbons 5—10 of I and others were in fairly good accord with the calculated values shown in Table II. Therefore AH_3 was concluded to be 6-hydroxy-2-(2-phenylethyl)chromone.

AH_4 (II), mp 89—90°C, $C_{18}H_{16}O_3$, was thought to be a methyl ether of I on the basis of its molecular formula and its IR spectrum, which showed no hydroxyl absorption. The ^{1}H-NMR spectrum showed the signal of a methoxyl group, and a *meta*-coupled proton (7.77 ppm, J=3.2 Hz) attached to C_5, suggesting the 6-methoxychromone structure. These data are supported by the similarity of the ^{13}C-NMR spectrum to that of I, except for the C_5 signal, which exhibited an upfield shift of about 3 ppm, and a methoxyl signal. Consequently AH_4 was concluded to be 6-methoxy-2-(2-phenylethyl)chromone.

AH_5 (III), $C_{19}H_{18}O_4$, mp 111—112°C, was suggested to be a chromone by the IR spectrum. The ^{1}H-NMR spectrum of AH_5 indicated the presence of two methoxyl groups and a chromone partial structure analogous to that of II. Therefore, one methoxyl group should be situated at the C_6 position of the chromone ring. In the phenyl skeleton the number of protons was decreased from five to four [6.94 (2H) and 7.21 ppm (2H)], indicating substitution by a methoxyl group. Since the ^{13}C-NMR spectrum showed a downfield shift at 1.6 ppm in the $C_{1'}$ signal of

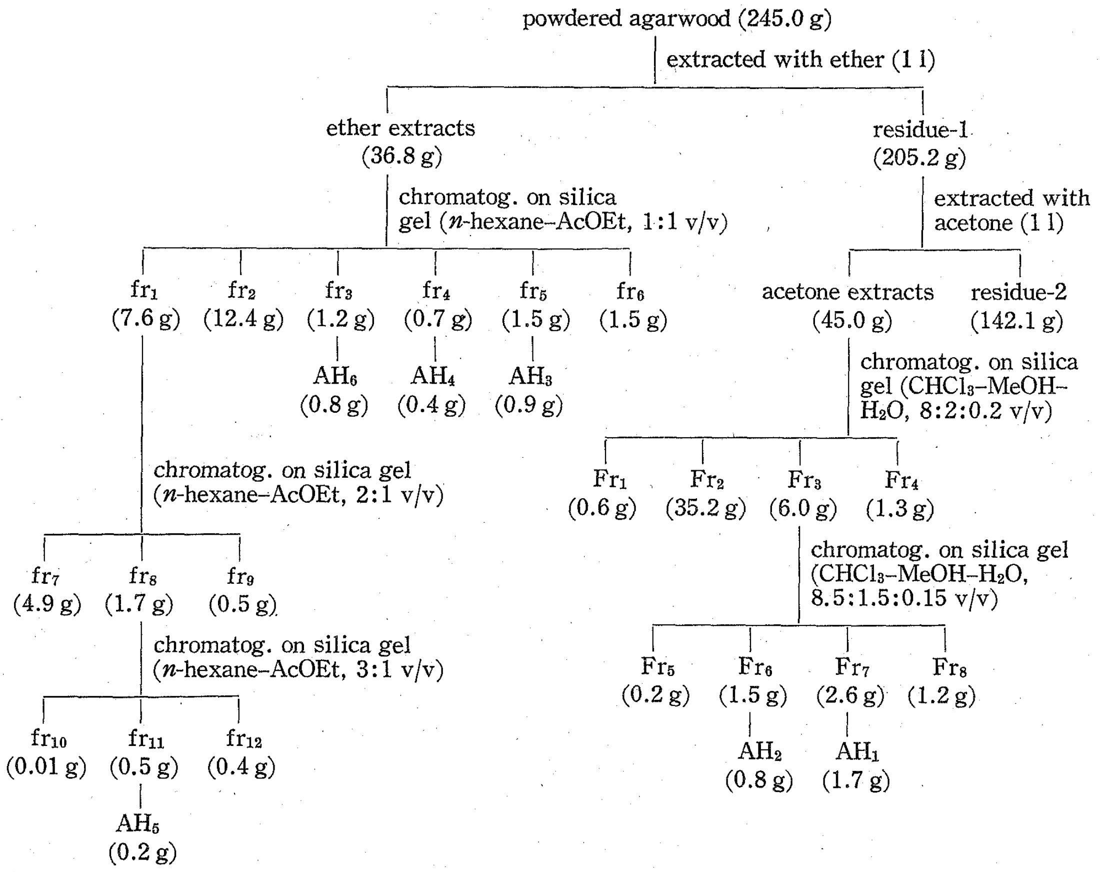
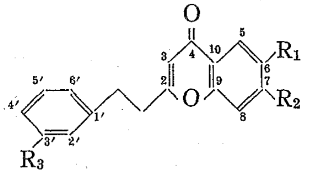


Chart 1

the phenylethyl group in comparison with data for I and II, it is considered that the methoxyl group is located at the *meta* position with respect to the 1′-carbon, and the other carbon signals showed excellent agreement with the calculated values as shown in Table II.[7] Accordingly AH_5 is considered to be 6-methoxy-2-[2-(3-methoxyphenyl)ethyl]chromone (III).

AH_6 (IV), $C_{19}H_{18}O_4$, mp 142—143°C, was indicated to be another dimethoxyl derivative of 2-(2-phenylethyl)chromone on the basis of the molecular formula, and IR and NMR spectra, but these spectra did not show signals due to a



I: $R_1=OH, R_2=R_3=H$
II: $R_1=OCH_3, R_2=R_3=H$
III: $R_1=R_3=OCH_3, R_2=H$
IV: $R_1=R_2=OCH_3, R_3=H$

Chart 2

Table I. ^1H-NMR Chemical Shifts (ppm from TMS) and Assignments

Carbon	AH_3	AH_4	AH_5	AH_6
3	6.30(s, 1H)	6.32(s, 1H)	6.34(s, 1H)	6.33(s, 1H)
5	8.04(t, J=1.4, 1H)	7.77(d, J=3.2, 1H)	7.77(d, J=3.2, 1H)	7.75(s, 1H)
7	7.48(m, 2H)	7.36(m, 2H)	7.36(m, 2H)	
8				7.05(s, 1H)
2′—6′	7.27(m, 5H)	7.27(m, 5H)	6.90(m, 2H) 7.21(m, 2H)	7.30(m, 5H)
CH_2CH_2	2.92(m, 4H)	2.92(m, 4H)	2.94(m, 4H)	2.95(m, 4H)
OMe		3.69(s, 3H)	3.68(s, 3H) 3.70(s, 3H)	3.76(s, 3H) 3.86(s, 3H)
OH	12.13(br, 1H)			

Table II. ^{13}C-NMR Chemical Shifts (ppm) of AH_3, AH_4, AH_5 and AH_6

Carbon[a]	AH_3	AH_4	AH_5	AH_6
2	168.32	168.34	168.56	167.66
3	109.37	109.61	109.64	109.84
4	177.79	177.35	177.56	176.84
5	108.96 (106.5)[b]	105.72 (102.0)	105.75 (102.0)	104.91 (104.0)
6	156.24 (155.5)	157.12 (158.8)	157.16 (158.8)	148.06 (148.0)
7	123.48 (123.5)	123.03 (123.4)	123.16 (123.4)	154.84 (154.2)
8	119.61 (119.0)	119.73 (118.4)	119.78 (118.4)	100.51 (100.1)
9	150.56 (150.2)	151.41 (151.7)	151.50 (151.7)	152.70 (152.6)
10	125.29 (125.1)	124.91 (124.0)	124.91 (124.0)	117.44 (118.1)
1′	140.61	140.57	142.20 (141.6)[c]	140.74
2′	128.89	128.91	114.59 (114.5)	128.97
3′	128.71	128.75	160.40 (160.2)	128.78
4′	126.70	126.76	112.34 (112.4)	126.77
5′	128.71	128.75	129.99 (129.8)	128.78
6′	128.89	128.91	121.03 (121.2)	128.97
CH_2CH_2	35.82	35.76	35.79	35.82
	32.95	32.94	33.04	33.07
6-OMe		55.61	55.69	55.89
7-OMe				56.27
3′-OMe			55.10	

a) The numbering system is indicated in the structural formulae.

b) Values in parentheses were calculated based on Table III (carbons 5—10).

c) Monomethoxy substituent effects,[7] applied to the data for AH_4. (carbons 1′—6′).

Table III. Hydroxy and Methoxy Substituent Increments (ppm) of Chromone Ring Carbons in Isoflavones[a]

Substituent(s) in position		Z_5[b]	Z_6	Z_7	Z_8	Z_9	Z_{10}	Data taken from compound(s)
a	5-OH	36.72	−15.09	2.39	−7.56	1.39	−10.61	**6**, **5**
b	6-OH	−18.85	29.69	−10.27	0.66	−6.44	0.09	**3**, **5**
c	7-OH	1.51	−10.21	28.81	−16.06	1.74	−7.35	**5**, **1**
d	8-OH	−3.43	2.21	−7.41	29.96	−6.89	−1.64	**8**, **5**
e	5-OMe	33.39	−18.49	−0.01	−7.12	1.39	−9.01	**7**, **2**
f	6-OMe	−23.28	32.96	−9.42	0.07	−4.87	−1.03	**4**, **7**
g	7-OMe	1.90	−10.69	29.91	−18.23	1.80	−5.87	**2**, **1**
h	8-OMe	−6.64	−4.02	−7.63	36.29	−7.56	0.43	**9**, **2**
								Increment sum
i	5,6-OH	17.87	14.60	−7.88	−6.90	−5.05	−10.52	a, b
j	5,7-OH	38.23	−25.30	31.20	−23.62	3.13	−17.96	a, c
k	5,8-OH	33.29	−12.88	−5.02	22.40	−5.50	−12.25	a, d
l	6,7-OH	−17.34	19.48	18.54	−15.40	−4.70	−7.26	b, c
m	6,8-OH	−22.28	31.90	−17.68	30.62	−13.33	−1.55	b, d
n	7,8-OH	−1.92	−8.00	21.40	13.90	−5.15	−8.99	c, d
o	5,6-OMe	10.11	14.50	−9.43	−0.05	−3.48	−10.04	e, f
p	5,7-OMe	35.29	−29.15	29.90	−25.35	3.19	−14.88	e, g
q	5,8-OMe	26.75	−22.48	−7.64	29.17	−6.17	−8.58	e, h
r	6,7-OMe	−21.38	22.27	20.49	−18.16	−3.07	−6.90	f, g
s	6,8-OMe	−29.92	28.94	−17.05	36.36	−12.43	−0.60	f, h
t	7,8-OMe	−4.74	−14.71	22.28	18.06	−5.76	−5.44	g, h

a) Increment values (a—h) were calculated based on the data for compounds **1**—**9** obtained here or quoted by Jha *et al.*[9]

b) Z_{5-10} indicate the increment values at C_{5-10} of the chromone ring, respectively.

1, isoflavone;[10] **2**, 7-methoxyisoflavone;[11] **3**, 6,7-dihydroxyisoflavone;[9] **4**, 6,7-dimethoxyisoflavone;[9] **5**, 7-hydroxy-4′-methoxyisoflavone;[11] **6**, 5,7-dihydroxyisoflavone;[12] **7**, 4′,5,7-trimethoxyisoflavone;[12] **8**, 4′,7,8-trihydroxyisoflavone;[9] **9**, 4′,7,8-trimethoxyisoflavone.[9]

phenethyl group bearing methoxyl functions. The ^{1}H-NMR spectrum of AH_6 exhibited signals of three singlet protons in the chromone ring. One of them, at 6.33 ppm, was assigned to the vinyl proton at C_3 as in I, II and III. The other two protons at 7.75 and 7.05 ppm are assumed to be located at the C_5 and C_8 positions of the chromone ring because each is a singlet, suggesting a 6,7-dimethoxyl substituted structure. The above results and the ^{13}C-NMR spectrum support the conclusion that AH_6 is 6,7-dimethoxy-2-(2-phenylethyl)chromone.

It is noteworthy that AH_3—AH_6 are chromones bearing a phenylethyl group at C_2, like the recently reported compound agarotetrol.[3] 2-Phenylethyl chromones have not been detected in normal tissues, except in the resinous parts of *Aquilaria malaccensis* LAM.[8] It is assumed that the presence of such constituents is specific to the process of resinification in Aquilaria plants.

The structures of AH_1 and AH_2 are under study.

Experimental

Melting points were determined on a micro melting point apparatus (Yanagimoto) and are uncorrected. UV spectra were obtained in EtOH with a Shimadzu UV-200s machine and IR spectra in KBr disks with a Shimadzu IR 27C spectrometer. ^{1}H and ^{13}C-NMR were taken in pyridine-d_5 solution on Varian A-60A and Varian CFT-20 machines, and chemical shifts are given in δ (ppm) with tetramethylsilane as an internal standard (s, singlet; d, doublet; t, triplet; m, multiplet; br, broad).

Isolation of AH_1—AH_6——Powdered agarwood (Jinkō, the middle grade on the market) from Kalimantan was extracted 3 times with ether under reflux for 3 h. The combined filtrate was concentrated to give a dark brown viscous extractive. Acetone extractive was obtained from the residue by the same procedure as above. These extracts were fractionated as shown in Chart 1. AH_3, AH_4, AH_5 and AH_6 were recrystallized from MeOH.

AH_3 (6-Hydroxy-2-(2-phenylethyl)chromone) (I)——Yellowish plates, mp 224—225°C, IR $\nu_{\max}$ cm^{-1}: 3275 (OH), 1655, 1638 (γ-pyrone), 1620, 1603, 1583, 1465, 1400 (aromatic ring). UV $\lambda_{\max}$ nm (ε): 207 (17660), 226 (23437), 240 (21302), 265 (6230), 328 (5727), 332 (5828). *Anal.* Calcd for $C_{17}H_{14}O_3$: C, 76.64; H, 5.29. Found: C, 76.64; H, 5.22.

AH_4 (6-Methoxy-2-(2-phenylethyl)chromone) (II)——Colorless plates, mp 89—90°C, IR $\nu_{\max}$ cm^{-1}: 1652, 1638 (γ-pyrone), 1604, 1597, 1562, 1480, 1434 (aromatic ring). UV $\lambda_{\max}$ nm (ε): 243 (10473), 273 (6060), 321 (6766), 324 (6472). *Anal.* Calcd for $C_{18}H_{16}O_3$: C, 77.12; H, 5.75. Found: C, 77.39; H, 5.84.

AH_5 (6-Methoxy-2-[2-(3-methoxyphenyl)ethyl]chromone) (III)——Colorless plates, mp 111—112°C, IR $\nu_{\max}$ cm^{-1}: 1655, 1640, (γ-pyrone), 1610, 1600, 1587, 1483, 1467, 1433 (aromatic ring). UV $\lambda_{\max}$ nm (ε): 244 (12532), 274 (10050), 321 (8512), 325 (8158). *Anal.* Calcd for $C_{19}H_{18}O_4$: C, 73.53; H, 5.85. Found: C, 73.30; H, 5.75.

AH_6 (6,7-Dimethoxy-2-(2-phenylethyl)chromone) (IV)——Colorless plates, mp 143—144°C, IR $\nu_{\max}$ cm^{-1}: 1658, 1638 (γ-pyrone), 1600, 1583, 1503, 1488, 1428 (aromatic ring). UV $\lambda_{\max}$ nm (ε): 212 (29247), 234 (26339), 278 (9674), 317 (10597). *Anal.* Calcd for $C_{19}H_{18}O_4$: C, 73.53; H, 5.85. Found: C, 73.34; H, 5.87.

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References and Notes

1) Part of this work presented at the 101st Annual Meeting of the Pharmaceutical Society of Japan, Kumamoto, April 1981.

2) K.R. Varma, M.L. Maheshwari and S.C. Bhattacharyya, *Tetrahedron*, **21**, 115 (1965); I.C. Jain and S.C. Bhattacharyya, *Tetrahedron Lett.*, **9**, 13 (1959); K.R. Varma and S.C. Bhattacharyya, *Tetrahedron*, **20**, 2927 (1964); M.L. Maheshwari, T.C. Jain, R.B. Bates and S.C. Bhattacharyya, *Tetrahedron*, **19**, 1079 (1963); M.L. Maheshwari, K.R. Varma and S.C. Bhattacharyya, *ibid.*, **19**, 1519 (1963).

3) E. Yoshii, T. Koizumi and T. Oribe, *Tetrahedron Lett.*, **41**, 3921 (1978).

4) K. Kafuku and N. Ichikawa, *Nippon Kagaku Zasshi*, **56**, 1155 (1935); N. Ichikawa and H. Yo, *ibid.*, **60**, 1247 (1939).

5) K. Sen and P. Bagchi, *J. Org. Chem.*, **24**, 316 (1959).

6) J.H. Noggle and R.E. Schirmer, "The Nuclear Overhauser Effect," Academic Press, New York, NY, 1971.

7) G.C. Levy, G.L. Nelson and J.D. Cargioli, *J. Chem. Soc., Chem. Commun.*, **1971**, 506; G.L. Nelson, G.C.

Levy and J.D. Cargioli, *J. Am. Chem. Soc.*, **94**, 3089 (1972).

8) Y. Shimada, T. Tominaga and S. Kiyosawa, The 101st Annual Meeting of the Pharmaceutical Society of Japan, Kumamoto, April 1981.

9) H.C. Jha, F. Zilliken and E. Breitmaier, *Can. J. Chem.*, **58**, 1211 (1980).

10) M.S. Chauhan and J.W. Still, *Can. J. Chem.*, **53**, 281 (1975).

11) A. Pelter, R.S. Ward and T.J. Gray, *J. Chem. Soc., Perkin Trans. 1*, **1976**, 2475.

12) A. Pelter, R.S. Ward and R.J. Buss, *J. Chem. Soc., Perkin Trans. 1*, **1978**, 666.