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Studies on the Constituents of *Swertia japonica*. IV.[1] Isolation and Structure of Xanthones[2]

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The chemical constituents of *Swertia japonica* Makino were studied. Three kinds of new natural xanthones, named norswertianin (I), swertianin (III) and methylswertianin (V), were isolated from the whole herb of this plant, together with desmethylbellidifolin (X) and bellidifolin (XII). I was identified as 1,3,7,8-tetrahydroxyxanthone. The direct isolation of I from natural source has never been published, although it has derived from decussatin (VII) and/or swertinin (VIII) by demethylation. The structure of III and V were shown to be 1,7,8-trihydroxy-3-methoxyxanthone and 1,8-dihydroxy-3,7-dimethoxyxanthone, respectively. At the same time, "swertianol" was revised the structure to 1,5,8-trihydroxy-3-methoxyxanthone, which was proved to be identical with bellidifolin.

Previously it was reported that C-glycosylflavonoids were isolated from the whole herb of *Swertia japonica* Makino (Japanese name "Senburi," Gentianaceae).[1,4]

The present paper deals with the isolation and structure of xanthone compounds in this plant. As shown in Chart 1 and described in detail in the experimental part, five xanthones were isolated in pure state on silica gel chromatography, *i.e.* compound A, mp 190°; compound B, mp 220°; compound C, mp 263°; compound D, mp 335°; and compound E, mp 317°, whose thin-layer chromatograms were shown in Fig. 1.

Compounds A, B and D were new natural xanthones, which would be named methylswertianin, swertianin and norswertianin, respectively.

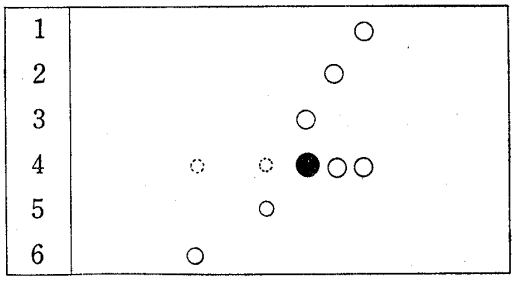


Fig. 1. Thin-Layer Chromatogram of Xanthones

plate: Silicagel G
solvent: toluene–ethylformate–formic acid (5:4:1)
color reagent: 5% Na_2CO_3
1, methylswertianin (yellow)
2, swertianin (dark brown)
3, bellidifolin (green→dark yellow)
4, xanthones of *Swertia japonica*
5, norswertianin
6, desmethylbellidifolin

Compound B (III), named swertianin, was obtained as yellow needles, mp 220°, and its analytical values suggested the formula $C_{14}H_{10}O_6$, exhibiting a positive ferric reaction. The infrared (IR) spectrum suggested the presence of hydroxyl groups (3450 cm^{-1}) and an α,β-unsaturated carbonyl group (1665 and 1645 cm^{-1} [KBr]), and the ultraviolet (UV) spectrum was characteristic of xanthone series giving the absorption maxima at 242, 269, 330 and 400 mμ (shoulder). It formed a triacetate, mp 184°/193° (double mp), $C_{20}H_{16}O_9$ (IV), on acetylation, whose IR spectrum showed the absence of hydroxyl group. Methylation of III with diazomethane yielded di-O-methylswertianin

1) Part III: M. Komatsu, T. Tomimori, Y. Makiguchi and K. Asano, *Yakugaku Zasshi*, **88**, 832 (1968).
2) This work was reported at the 88th Annual Meeting of Pharmaceutical Society of Japan, Tokyo, April, 1968.
3) Location: *No. 34–1, Takata 3-chome, Toshimaku, Tokyo.*
4) M. Komatsu, T. Tomimori and M. Ito, *Chem. Pharm. Bull.* (Tokyo), **15**, 263 (1967); M. Komatsu, T. Tomimori and Y. Makiguchi, *ibid.*, **15**, 1567 (1967).

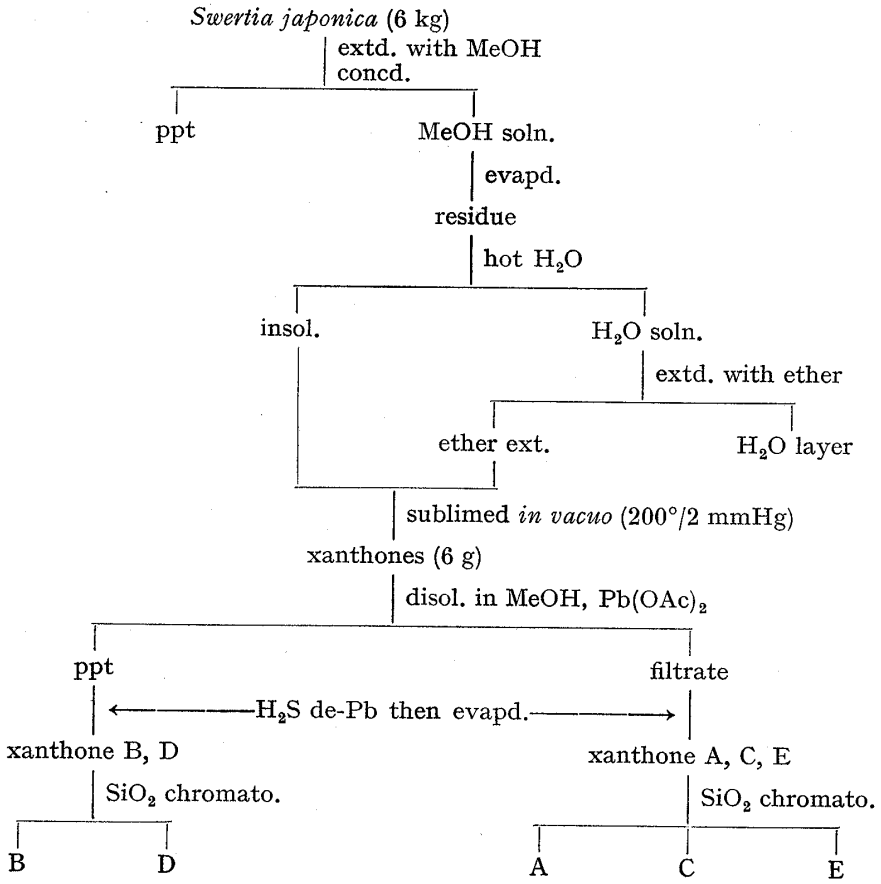


Chart 1. Extraction and Isolation of Xanthones

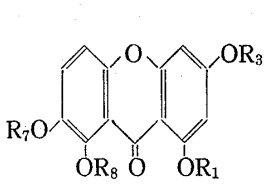
(VII), mp 150°, $C_{16}H_{14}O_6$, with a positive ferric reaction. Demethylation of III with hydriodic acid gave tetrahydroxyxanthone, mp 335°, $C_{13}H_8O_6$, which was identical with compound D (I) by comparison of itself and its tetraacetate, mp 225°, $C_{21}H_{16}O_{10}$ (II). III gave a nuclear magnetic resonance (NMR) spectrum (Table I) showing the presence of one methoxyl at τ 6.12 and hydroxyl groups at τ −1.9, −1.7 and 0. These facts indicated that III was a trihydroxy–monomethoxyxanthone. The absence of C_1 or C_8 carbonyl–deshielded protons (which should occur at about τ 2[5]) indicates that III is oxygenated in the 1 and 8 positions. The presence of a 1,8-dihydroxy system is strongly suggested by the two hydrogen–bonded hydroxyl protons at τ −1.9 and τ −1.7. In such a system, methylation of one hydrogen–bonded hydroxyl group would be expected to increase the extent of hydrogen bonding of the other and thus produce a downfield shift of its signal.[5d] This is found to be the case of di-O-methylswertianin (VII) where the one remaining hydrogen–bonded hydroxyl proton has shifted downfield to τ −2.7, a value of which is in accord with those observed for 1-hydroxy-8-methoxyxanthones such as 1-hydroxy-3,8-dimethoxyxanthone (τ −3.3) and bellidifolin dimethylether (τ −2.7).[5d]

Coupled doublets centred at τ 2.72, τ 3.15 (J=9 cps) and τ 3.46, τ 3.68 (J=2.5 cps) represent two pair of aromatic protons which are assigned to *"ortho"* and *"meta"* proton pairs, respectively; thus the only oxidation patterns tenable for III are 1,3,5,8 and 1,3,7,8.

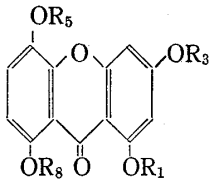
That a methoxyl group must be placed at position 3 is proved in the following ways. (a) Compound D (I) dissolves readily in sodium carbonate solution,[5d,5e,6] while III does not. (b) The long wave length absorption in the UV spectrum of I shifted bathochromically by

5) *a*) C.T. Mathis and J.H. Goldstein, *Spectrochimica Acta*, **20**, 871 (1964); *b*) E.D. Burling, A. Jefferson and F. Scheinmann, *Tetrahedron*, **21**, 2653 (1965); *c*) L. Crombie and D. Whiting, *Tetrahedron Letters*, **1962**, 801; *d*) K.R. Markham, *Tetrahedron*, **20**, 991 (1964); *e*) *Idem*, *ibid.*, **21**, 1449, 3687 (1965).

6) G.V. Rao and T.R. Seshadri, *Proc. Indian Acad. Sci.*, **37A**, 710 (1953).



	R_1	R_3	R_7	R_8
I :	H	H	H	H
II :	Ac	Ac	Ac	Ac
III :	H	Me	H	H
IV :	Ac	Me	Ac	Ac
V :	H	Me	Me	H
VI :	Ac	Me	Me	Ac
VII :	Me	Me	Me	H
VIII :	Me	Me	H	H
IX :	H	H	Me	H



	R_1	R_3	R_5	R_8
X :	H	H	H	H
XI :	Ac	Ac	Ac	Ac
XII :	H	Me	H	H
XIII :	Ac	Me	Ac	Ac
XIV :	H	H	Me	H
XV :	H	Me	Me	Me

Chart 2

TABLE I. Nuclear Magnetic Resonance Spectra of Xanthones

	Solv.	Hydroxyl protons	Acetyl protons	Methoxyl protons	Aromatic protons $J=2.5$ cps	Aromatic protons $J=9$ cps
I	c	−1.9, −1.7, −1.2, 0.7			3.82, 3.72	3.21, 2.68
II	a		7.59(6H), 7.70(6H)		3.23, 2.83	2.73, 2.53
III	c	−1.9, −1.7, 0		6.12(3H)	3.68, 3.46	3.15, 2.72
IV	a		7.70(3H), 7.58(6H)	6.12(3H)	3.48, 3.29	2.73, 2.55
V	b	−1.9, −1.7		6.10(6H)	3.80, 3.70	3.25, 2.70
VI	a		7.57(3H), 7.55(3H)	6.11(3H), 6.13(3H)	3.75, 3.57	3.50, 3.30
VII	c	−2.7		6.10(3H), 6.15(3H) 6.12(3H)	3.81, 3.60	3.10, 2.60
IX	c	−1.9, −1.7		6.12(3H)	3.70, 3.56	3.00, 2.50
X	c	−1.9, −1.2, 0			3.81, 3.60	3.42, 2.80
XI	a		7.79(3H), 7.66(9H)		3.43, 3.02	3.33, 2.99
XII	c	−1.9, −1.1, 0.2		6.12(3H)	3.68, 3.48	3.40, 2.78
XIII	a		7.59(9H)	6.12(3H)	3.47, 3.35	3.14, 2.66
XIV	c	−1.8, −1.2, 0		6.13(3H)	3.76, 3.60	3.30, 2.90
XV	a	−2.6		6.05(3H), 6.10(3H) 6.15(3H)	3.73, 3.48	3.40. 2.90

NMR spectra were measured at 60 Mcps. Chemical shifts were expressed in τ value from TMS.
Deuterium oxide was added to identify hydroxyl groups.
solvent: a) deuterochloroform
b) deuterochloroform-deuterodimethylsulfoxide (1:2)
c) deuterodimethylsulfoxide

TABLE II. Color Reaction of *ortho*- and *para*-Diphenol

Compound	Lead acetate (in MeOH)	Ammonium molybdate	Cobalt reagent[a] $[Co(NH_3)_5Cl]Cl_2$ (in hydr.EtOH)[b]	Tollens reagent
I	orange precipitate	dark brown precipitate	dark brown precipitate	(+)
III	orange precipitate	dark brown precipitate	brown precipitate	(+)
V	yellow (−)	(−)	(−)	(−)
IX	yellow (−)	(−)	(−)	(−)
X	yellow (−)	(−)	dark brown precipitate	(+)
XII	yellow (−)	(−)	dark brown precipitate	(+)
XIV	yellow (−)	(−)	(−)	(−)

a) M. Hasegawa, "*Zikken Kagaku Koza*," Vol. 22, ed. by The Chemical Society of Japan, Maruzen Co., Ltd., Tokyo, 1958, p. 291
b) heated at below 60° on a water bath

28 mμ in the presence of sodium acetate whilst no change was observed in the spectrum of III.[5e,7] These reduce the structural possibilities of III to two only, *i.e.* 1,7,8-trihydroxy-3-methoxyxanthone and 1,5,8-trihydroxy-3-methoxyxanthone (bellidifolin). UV spectrum of III is very similar to those of 1,3,7,8-oxygenated xanthones,[5e,7] and III differ spectrally (IR and UV) and thin-layer chromatographycally from authentic bellidifolin.[8] Further, III containing an *ortho*-dihydroxy system was proved by the color reactions (Table II).

From these results the structure of III was established as 1,7,8-trihydroxy-3-methoxyxanthone. At the same time, compound D (I) should therefore be 1,3,7,8-tetrahydroxyxanthone containing the same oxidation pattern as swertinin (VIII) and decussatin (VII), both of which have been isolated from *Swertia decussata*.[9]
Although I has not yet been found in nature, it has been obtained by Shah, *et al.*[9] through demethylation of swertinin and decussatin. Hence, we propose the name norswertianin to compound D.

Compound A (V), named methylswertianin, was obtained as pale yellow needles, mp 190°, $C_{15}H_{12}O_6$. It forms diacetate, mp 210°, $C_{19}H_{16}O_8$ (VI), and has UV spectrum very similar to those of I and III, indicating that the 1,3,7,8-oxidation pattern is present. This was confirmed by formation of di-O-methylswertianin (VII) on methylation and norswertianin (I) through demethylation. The NMR spectrum (Table I) shows the presence of two hydroxyl, four aromatic and six methoxyl protons. V should therefore be norswertianin-dimethylether. The 1,8-dihydroxy system is still present (τ −1.9, τ −1.7), indicating that two methoxyl groups must be placed at 3 and 7 positions. In addition, following data support the structure. (a) Color reactions of orthodiphenol are negative. (b) Unlike norswertianin, V was insolble in sodium carbonate solution. (c) The presence of a 3-methoxyl group in V is also confirmed by the comparison of its UV spectra with those in the presence of sodium acetate (no change). From the foregoing results, the structure 1,8-dihydroxy-3,7-dimethoxyxanthone was assigned to methylswertianin.

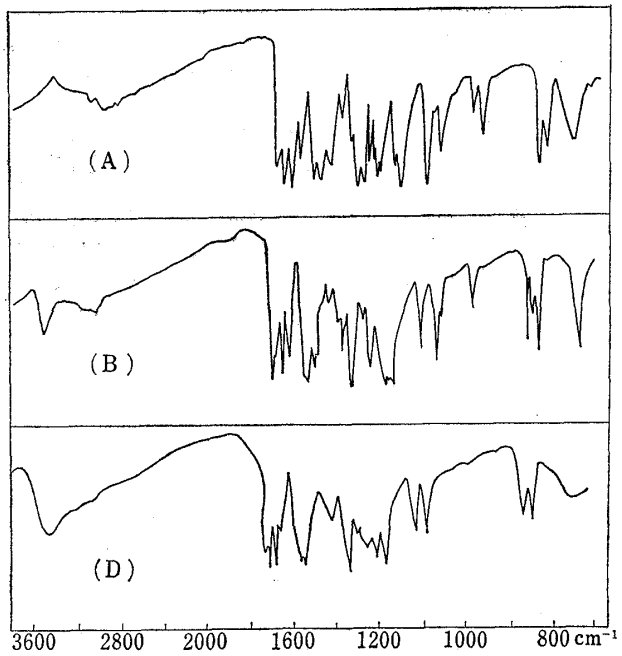


Fig. 2. Infrared Absorption Spectra of A, B and D (in KBr)
(A): methylswertianin (B): swertianin (D): norswertianin

Since compound A, B and D were new natural xanthones, the syntheses of them were attempted. 2-Hydroxy-5,6-dimethoxyacetophenone[10] was prepared from resorcinol and oxidized by a modified iodoform reaction[5e,11] to 2-hydroxy-5,6-dimethoxybenzoic acid, mp 80°, which has already been prepared by the sodium hypochlorite oxidation of the acetophenone by Dalal, *et al.*[12] but not reported in detail. Condensation of this acid with phloroglucinol in the presence of phosphoryl chloride and zinc chloride by Shah's method[12,13], gave a 1:1 mixture of 1,3,8-trihydroxy-7-methoxyxan-

7) R.C. Shah, A.B. Kulkarni and S.R. Dalal, *J. Sci. Ind. Res.*, **13B**, 175 (1954).
8) The sample was furnished through the courtesy of Dr. K.R. Markham.
9) S.R. Dalal, S. Sethna and R.C. Shah, *J. Indian Chem. Soc.*, **30**, 457 (1953).
10) W. Baker, *J. Chem. Soc.*, **1939**, 956.
11) L.C. King, *J. Am. Chem. Soc.*, **66**, 894 (1944).
12) S.R. Dalal and R.C. Shah, *Chem. Ind.* (London), **1957**, 140.
13) P.K. Grover, G.D. Shah and R.C. Shah, *J. Chem. Soc.*, **1955**, 3982.

thone (IX) and 1,3,8-trihydroxy-5-methoxyxanthone (XIV). The melting point for the former (300°[5e,12]) and its spectra (UV and NMR[5e]) agreed closely with those of the compound. The latter was proved to be identical with isobellidifolin by a direct comparison with the authentic sample[8].

Subsequently, IX being selectively methylated, gave 1,8-dihydroxy-3,7-dimethoxyxanthone which was identical with natural methylswertianin (V). IX was also demethylated to 1,3,7,8-tetrahydroxyxanthone. This was identical with norswertianin (I) and selective methylation gave 1,7,8-trihydroxy-3-methoxyxanthone, identical with swertianin (III).

Compound C (XII) was obtained as yellow needles, mp 263°, $C_{14}H_{10}O_6$, exhibiting a positive ferric reaction. The NMR spectrum (Table I) showed the presence of two hydrogen-bonded hydroxyl, a non-chelated one (broad peak), four aromatic and three methoxyl protons. The UV spectrum of XII was found closely resemble to that of isobellidifolin (XIV), suggesting that the 1,3,5,8-oxidation pattern was present. Demethylation of XII with hydriodic acid gave compound E (X), mp 317°, $C_{13}H_8O_6$, which was identical with desmethylbellidifolin by comparison of itself and its tetracetate (XI), mp 244°, $C_{21}H_{16}O_{10}$, prepared by the demethylation of isobellidifolin XII should, therefore, be 1,3,5,8-tetrahydroxyxanthone-monomethyl ether. XII also having 3-methoxyl group was confirmed by the same method as in the case of swertianin.

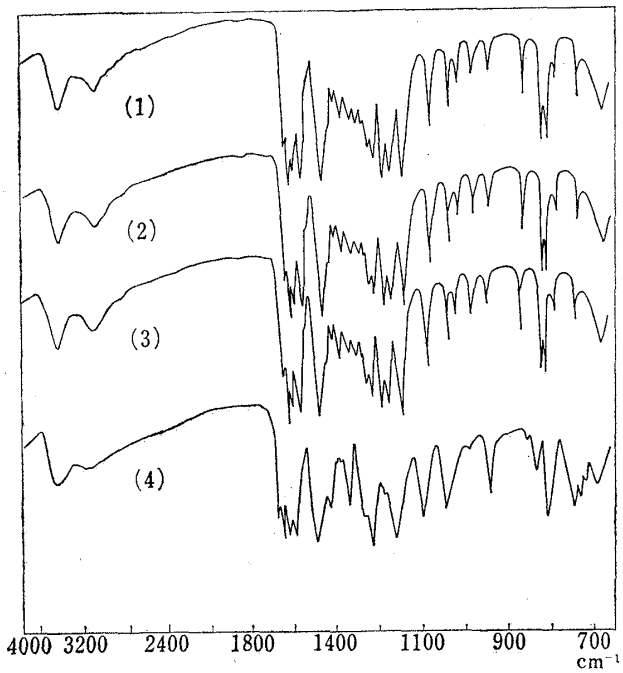


Fig. 3. Infrared Absorption Spectra of 1, 2,3 and 4 (inKBr)

(1): bellidifolin (2): compound C
(3): "swertianol" (4): isobellidifolin

From the foregoing findings, XII may be identical with bellidifolin, isolated from *Gentiana bellidifolia* by Markham.[5d] Finally, mixed melting point determination, co-chromatography on silica gel plates, and comparison of UV and IR spectra with authentic specimen[8] established the identity of XII with bellidifolin. The triacetate (XIII) did not depress the melting point on admixture with authentic bellidifolin triacetate.

In 1942, "swertianol" was isolated from the whole herb of *Swertia japonica* by Asahina, *et al.*,[14] who proposed the two possibilities of structure which were given 1,3,8-trihydroxy-5-methoxyxanthone and 1,3,5-trihydroxy-8-methoxyxanthone. Subsequently, Nakaoki, *et al.*[15] isolated "swertianol", identified with Asahina's sample, from *Swertia diluta* var. *tosaensis* which they claimed possessed structure XIV. The presence of a 5-methoxyl group in "swertianol" was determined by color reaction (negative quinol test)[16] without other physical data and for this reason the exact structure has been considered in doubt.[5d,17]

On the other hand, isobellidifolin (XIV) was isolated together with bellidifolin from *Gentiana bellidifolia* by Markham,[5e] who proposed the same structure XIV for isobellidifolin.

14) Y. Asahina, J. Asano and Y. Ueno, *Yakugaku Zasshi*, **62**, 22 (1942).
15) T. Nakaoki and Y. Hida, *Yakugaku Zasshi*, **63**, 554 (1943).
16) The absence of paradiphenol was tested by cobalt reagent at room temperature by Asahina, *et al.*,[14] but it has been found barely discernible positive in a negative in this conditions (Table II).
17) J.C. Roberts, *Chem. Rev.*, **61**, 592 (1961).

In order to determine whether or not "swertianol" is identical with isobellidifolin, we have compared "swertianol"[18] with isobellidifolin. In consequence, it was found that "swertianol" differed IR spectrally (Fig. 3) and thin-layer chromatographycally from isobellidifolin. Moreover, "swertianol" could be distinguished from isobellidifolin in the following ways. (a) "Swertianol" gave a positive paradiphenol test, while isobellidifolin did not. (b) The long wave length absorption in the UV spectrum of isobellidifolin shifted bathochromically by 26 mμ in the presence of sodium acetate whilst no change was observed in "swertianol", suggesting that "swertianol" had a 3-methoxyl group. (c) Isobellidifolin dissolves readily in sodium carbonate solution, while "swertianol" does not.

These data show that "swertianol" may be identical with bellidifolin. Consequently, a comparison of "swertianol" with bellidifolin in mixed melting point determination, co-chromatography on silica gel plate, UV and IR spectra established their identity.

Thus, it has now become evident that xanthone compounds in the whole herb of *Swertia japonica* Makino consisted of 1,3,5,8- and 1,3,7,8-oxygenated xanthones, and it is of interest to support the hypothesis that the 5 and 7 positions in xanthones are biogenetically equivalent if ring closure of the central γ-pyrone ring take place at the ether oxygen.

Experimental

All melting points were uncorrected. UV spectra were measured after Jurd[19], using a Hitachi Recording Spectrophotometer EPS-2U type. IR spectra were determined on KBr discs using a JASCO DS-301 Spectrophotometer. NMR spectra were measured on a Hitachi Perkin-Elmer Spectrometer (Model R-20) and on a Varian A-60 Spectrometer. The chemical shifts were expressed in τ value from TMS. Thin-layer chromatograhy was carried out on silica gel G with toluene–ethyl formate–formic acid (5:4:1) as solvent. Solubility of xanthones were tested simultaneously at 20° in 10% Na_2CO_3 aqueous solution. III, V and XII remained insoluble for 10 min whilst I, IX, X and XIV were immediately soluble.

Extraction and Isolation of Xanthones——The dried whole herb of *Swertia japonica* Makino (6 kg) was treated as shown in Chart 1. A mixture of compound B and D was chromatographed on a column of silica gel, using benzene containing 10% AcOEt as an eluant, and each fractions were checked by TLC. The faster-moving fraction consisted of compound B, and the slower-moving fraction consistecd of compoud D, respectively. A mixture of compound A, C and E was submitted to column chromtography of silica gel and eluted with benzene containing 5% AcOEt. After elution of compound A with the solvent, successive elution of the chromatogram with benzene containing 15% AcOEt gave compound C and compound E in turn. Yield: A (150 mg), B (200 mg), C(5 g), D (50 mg), E (40 mg).

Compound B (Swertianin) (III)——III was recrystallized from AcOEt to yellow needles, mp 220°. $FeCl_3$ (+). UV λ_{max}^{EtOH} mμ (log ε): 242 (4.40), 269 (4.48), 314 (sh.) (4.10), 330 (4.12), 400 (sh.) (3.70). No change was observed when the spectrum was determined in the presence of NaOAc. *Anal.* Calcd. for $C_{14}H_{10}O_6$: C, 61.32; H, 3.68. Found: C, 61.52; H, 3.60.

Tri-O-acetylswertianin (IV): III (60 mg) was dissolved in 1 ml of pyridine, and 1 ml of Ac_2O was added and heated at 100° for 2 hr. The mixture was poured into ice water. The product was recrystallized from MeOH to obtain colorless needles, mp 184°/193°. Yield, 30 mg, $FeCl_3$ (−). IR cm^{-1} (KBr): 1770 ($COCH_3$); 1660, 1638 (conjugated CO); 1595, 1572, 1480 (aromatic C=C). *Anal.* Calcd. for $C_{20}H_{16}O_9$: C, 60.00; H, 4.03. Found: C, 59.72; H, 3.94.

Di-O-methylswertianin (VII): A dry ethereal solution of CH_2N_2 generated from nitrosomethylurethane (4 ml) was added to a MeOH solution of III (40 mg) at 2°. The mixture was allowed to stand overnight. After removal of the solvent, the residue was recrystallized from AcOEt, forming yellow needles (22 mg), mp 150°. $FeCl_3$ (+). UV λ_{max}^{EtOH} mμ (log ε): 240 (4.56), 260 (4.64), 315 (4.16), 380 (sh.) (3.68). *Anal.* Calcd. for $C_{16}H_{14}O_6$: C, 63.57; H, 4.67. Found: C, 63.62; H, 4.59.

Demethylation of III: A mixture of III (20 mg), PhOH (1 ml) and HI (1 ml, d=1.7) was gently boild under reflux for 7 hr. After being cooled, the reaction mixture was poured into 1% $NaHSO_3$ solution. A yellow deposit was obtained which on recrystallization from 40% EtOH gave yellow needles, mp 335°. $FeCl_3$ (+). Admixture with compound D did not depress the melting point. It was also spectrally (IR

18) The sample, has been preserved at the Faculty of Pharmaceutical Sciences, Tokyo University, was furnished through the courtesy of Dr. O. Tanaka.

19) *a*) L. Jurd and R.M. Horowitz, *J. Org. Chem.*, **22**, 1618 (1957); *b*) L. Jurd, *Arch. Biochem. Biophys.*, **63**, 376 (1956).

and UV) and chromatographycally (TLC) identical with compound D. Acetylated with Ac_2O–pyridine, it gave an acetate which was recrystallized from MeOH in colorless needles, mp 225°. The acetate was identical with tetra-O-acetyl compound D by the comparison of mixed mp and IR spectra.

Compound A (Methylswertianin) (V)——V was recrystallized from MeOH to pale yellow needles, mp 190°. $FeCl_3$ (+). UV λ_{max}^{EtOH} mμ (log ε): 240 (4.42), 265 (4.48), 312 (sh.) (4.12), 330 (4.14), 386 (sh.) (3.68). No changes were observed when the spectrum was determined in the presence of NaOAc or NaOAc–H_3BO_3.

Di-O-acetylmethylswertianin (VI): Ac_2O–pyridine treatment of V produced a diacetate which was recrystallized from MeOH in colorless needles, mp 210°. *Anal.* Calcd. for $C_{19}H_{16}O_8$: C, 61.29; H, 4.33. Found: C, 61.35; H, 4.40. Demethylation of V: A mixture of V (20 mg), PhOH (0.5 ml) and HI (1 ml, d=1.7) was boiled under reflux for 7 hr. The reaction mixture was worked up as usual and recrystallized from 40% EtOH to obtain yellow needles, mp 335°, which was identified with compound D by TLC, mixed fusion, UV and IR. Methylation of V:V (20 mg) in MeOH (20 ml) was left overnight with an excess of ethereal CH_2N_2 at 5°. The product was recrystallized from AcOEt, giving yellow needles, mp 150°, which was identical with di-O-methylswertianin (VII) by mixed fusion and UV.

Compound D (Norswertianin) (I)——I was recrystallized from 40% EtOH to yellow needles, mp 335°. $FeCl_3$(+). UV λ_{max}^{EtOH} mμ (log ε): 239 (4.42), 267 (4.50), 332 (4.10), 392 (sh.) (3.70). UV $\lambda_{max}^{EtOH-NaOAc}$ mμ (log ε): 270 (4.40), 360 (4.30). *Anal.* Calcd. for $C_{13}H_8O_6$: C, 60.01; H, 3.10. Found: C, 59.95; H, 3.21.

Tetra-O-acetylnorswertianin (II): I on treatment with boiling Ac_2O–pyridine yielded an acetate, which was recrystallized from MeOH in colorless needles, mp 225°.

IR cm^{-1} (KBr): 1770 ($COCH_3$); 1665, 1630 (conjugated CO); 1600, 1578, 1490 (aromatic C=C). *Anal.* Calcd. for $C_{21}H_{16}O_{10}$: C, 58.88; H, 3.77. Found: C, 58.77; H, 3.86.

Methylation of I: I in MeOH was left overnight with an excess of ethereal CH_2N_2. The product was worked up as usual and recrystallized from AcOEt to obtain yellow needles, mp 150°, which was identical with di-O-methylswertianin (VII) by mixed fusion and UV.

Compound C (Bellidifolin) (XII)——Yellow needles, mp 263° (from EtOH). $FeCl_3$ (+). XII was identified with authentic specimen[8] by TLC, mixed fusion, IR and UV. XII was also identical with "swertianol"[18] by the comparison of TLC, mixed fusion, IR and UV. UV λ_{max}^{EtOH} mμ (log ε): 255 (4.22), 279 (4.10), 335 (3.90), 400 (sh.) (3.70). No changes were observed when the spectrum was determined in the presence of NaOAc or NaOAc–H_3BO_3. *Anal.* Calcd. for $C_{14}H_{10}O_6$: C, 61.32; H, 3.68. Found: C, 61.26; H, 3.92.

Tri-O-acetylbellidifolin (XIII): XII was acetylated with Ac_2O–pyridine (2 hr at 100°). The product was recrystallized from MeOH to obtain colorless needles, mp 238°, which was identical with authentic bellidifolin triacetate by mixed fusion. IR cm^{-1} (KBr): 1768 ($COCH_3$); 1650, 1634 (conjugated CO); 1615, 1590, 1568, 1480 (aromatic C=C). *Anal.* Calcd. for $C_{20}H_{16}O_9$: C, 60.00; H, 4.03. Found: C, 59.72; H, 3.95.

Demethylation of XII: A mixture of XII(200 mg), PhOH(4 ml) and HI(5 ml, d=1.7) was boiled gently under reflux for 7.5 hr. The product was worked up as usual and recrystallized from EtOH–H_2O to obtain yellow needles, mp 317°, which was identified with compound E by mixed fusion, TLC, IR and UV. Its acetate, prepared by the Ac_2O–pyridine method, melted at 244°, which was identical with tetra-O-acetyl-compound E by mixed fusion and IR. Di-O-methylbellidifolin (XV): XII (40 mg) in MeOH (50 ml) was left overnight with an excess of ethereal CH_2N_2 at 5°. The product (40 mg) crystallized from AcOEt as yellow needles, mp 205°. $FeCl_3$(+). UV λ_{max}^{EtOH} mμ (log ε): 254(4.4), 278(4.2), 300(sh.) (3.8), 335(4.0). IR cm^{-1} (KBr): 3200—2600 (OH); 1660 (conjugated CO); 1614, 1595, 1575, 1495 (aromatic C=C). *Anal.* Calcd. for $C_{16}H_{14}O_6$: C, 63.57; H, 4.67. Found: C, 63.77; H, 4.38.

Compound E (Desmethylbellidifolin) (X)——Yellow needles, mp 317° (from EtOH–H_2O). $FeCl_3$(+). UV λ_{max}^{EtOH} mμ (log ε): 255 (4.35), 279 (4.20), 337 (4.10), 390 (sh.) (3.70). UV $\lambda_{max}^{EtOH-NaOAc}$ mμ (log ε): 250 (4.23), 273 (4.21), 362 (4.23). IR cm^{-1} (KBr): 3400 (OH); 1670, 1645 (conjugated CO); 1620, 1595, 1508 (aromatic C=C). *Anal.* Calcd. for $C_{13}H_8O_6$: C, 60.01; H, 3.10. Found: C, 60.22; H, 3.15.

Tetra-O-acetyldesmethylbellidifolin (XI): Ac_2O-pyridine treatment of X yielded a tetraacetate, which was recrystallized from AcOH to obtain colorless needles, mp 244°. $FeCl_3$(−).

IR cm^{-1} (KBr); 1775 ($COCH_3$); 1660, 1634 (conjugated CO); 1615, 1578, 1490 (aromatic C=C). *Anal.* Calcd. for $C_{21}H_{16}O_{10}$: C, 58.88; H, 3.77. Found: C, 58.49; H, 4.04.

2-Hydroxy-5,6-dimethoxybenzoic Acid——2-Hydroxy-5,6-dimethoxyacetophenone[10] (8.4 g or 0.043 mole) in pyridine (84 ml) was added to a solution of I_2 (11 g or 0.045 mole) and left at room temperature for 3 hr. After a further 2 hr at 100° the solution was evaporated to dryness *in vacuo* and 3% NaOH solution (300 ml) was added. The alkaline solution was heated for 1 hr on a steam-bath and acidified with dil. HCl, and then extracted with ether. The ether extract was fractionated by the usual method into an acidic fraction. The crude acid was chromatographed on a column of silica gel, using benzene–$CHCl_3$ (4:1) as an eluant. After removal of the solvent, the white solid obtained was recrystallized from MeOH, giving colorless needles (4 g), mp 79° (lit.[12] mp 79°).

Isobellidifolin (XIV)——2-Hydroxy-5,6-dimethoxybenzoic acid (3 g) was mixed with dried phloroglucinol (2.5 g), $POCl_3$ (30 ml) and freshly fused $ZnCl_2$ (9 g) and heated at 80—90° for 2 hr. After being cooled,

20) Color reagent; 5% Na_2CO_3: isobellidifolin (yellow) *cf.* bellidifolin (green→dark yellow).

the reaction mixture was poured into ice water. The precipitate obtained was triturated with hot AcOEt. After removal of the solvent, the brown powder obtained was sublimed *in vacuo*, giving the mixture of two xanthones (0.5 g). TLC on silica gel (developer: benzene–AcOEt=1:1) afforded two spots at *Rf* 0.35 and 0.5. This material was submitted to column chromatography of silica gel and eluted with benzene containing 10% AcOEt. The faster-moving fractions indicating only one spot at *Rf* 0.5 on TLC, were combined and evaporated to dryness, giving yellow powdery crystalls, which was recrystallized from EtOH as yellow needles, mp 263°. $FeCl_3$(+). UV λ_{max}^{EtOH} mμ (log ε): 253 (4.3), 277 (4.2), 336 (4.1), 380 (sh.) (3.7). UV $\lambda_{max}^{EtOH\text{-}NaOAc}$ mμ (log ε): 247 (4.20), 274 (4.15), 362 (4.20). The identity of XIV with authentic isobellidifolin[8] was proved by TLC[20], UV and IR.

Demethylation of XIV: XIV was demethylated in 70% yield by above-mentioned procedure using HI, giving desmethylbellidifolin, mp 317°, which was identical with compound E by mixed fusion, TLC, IR and UV.

1,3,8-Trihydroxy-7-methoxyxanthone (IX)——The slower-moving fractions in above-mentioned column chromatography indicating only one spot at *Rf* 0.35 on TLC, were combined and evaporated to dryness, giving yellow powder, which was recrystallized from AcOEt as yellow needles, mp 300° (lit.[12] mp 300°). $FeCl_3$(+). UV λ_{max}^{EtOH} mμ (log ε): 238 (4.45), 262 (4.50), 334 (4.20), 380 (sh.) (3.67). UV $\lambda_{max}^{EtOH\text{-}NaOAc}$ mμ (log ε): 263 (sh.) (4.41), 270 (4.43), 361 (4.40). IR cm^{-1} (KBr): 3360 (OH); 1660, 1642 (conjugated CO); 1614, 1578, 1515, 1480 (aromatic C=C). *Anal.* Calcd. for $C_{14}H_{10}O_6$: C, 61.32; H, 3.68. Found: C, 61.29; H, 3.97.

Demethylation of IX: IX was demethylated by above-mentioned procedure using HI, giving yellow needles, mp 335°, which was identified with compound D by TLC, mixed fusion, UV and IR.

Partial Methylation of IX——IX (52 mg) in dry acetone (20 ml) was refluxed with dimethyl sulfate (30 mg) and $NaHCO_3$ (1 g) for 15 hr. After filtration and removal of the solvent, the residue obtained was sublimed *in vacuo* and recrystallized from MeOH, giving pale yellow needles, mp 190°, which was spectrally (IR and UV) and chromatographycally (TLC) identical with compound A.

Partial Methylation of I——I (50 mg), prepared by synthesis, was treated as described above for the partial methylation of IX, giving 1,7,8-trihydroxy-3-methoxyxanthone, mp 220°. Admixture with compound B did not depress the melting point and the IR and UV spectra were also found to be superimposable with those of compound B.

Partial Methylation of X——X (52 mg), prepared by synthesis, was partially by methylated by above-mentioned procedure, forming bellidifolin, which was identified with compound C by mixed fusion, TLC, IR and UV.

On acetylation with Ac_2O–pyridine, it gave an acetate, mp 238°, which was also identical with tri-O-acetylbellidifolin by the comparison of mixed fusion and IR spectra.

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