# Constituents of *Iva* Species. V. Isolation, Structure, and Synthesis of Nevadensin, a New Flavone from *Iva Nevadensis* M. E. Jones and *Iva Acerosa* (Nutt.) Jackson[1,2]

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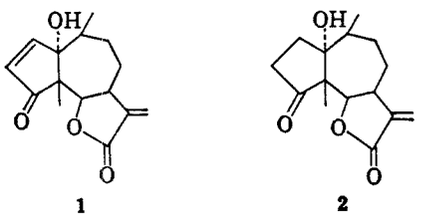
A new flavone nevadensin has been isolated from *Iva nevadensis* M. E. Jones and *Iva acerosa* (Nutt.) Jackson and its structure has been established as 5,7-dihydroxy-4′,6,8-trimethoxyflavone by synthesis. Identification of nevadensin and pectolinarigenin, also found in *I. nevadensin*, was complicated by unexpected spectral properties which depart from generally accepted rules for structural diagnosis of 7-hydroxyflavonoids. The pseudoguaianolides parthenin and coronopilin were also isolated from *I. nevadensis; I. acerosa* furnished coronopilin.

As part of our inquiry into possible connections between genera related to *Ambrosia* and *Parthenium*, we have undertaken a chemical survey of the genus *Iva*. In the present paper we report the results of our examination of *Iva nevadensis* M. E. Jones, a somewhat uncommon species whose distribution is limited to western Nevada and adjacent California, and *Iva acerosa* (Nutt.) Jackson (copper weed),[6] which is widely distributed in the western United States.



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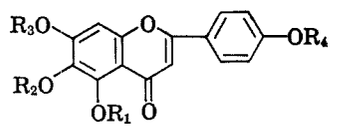

**3a**, $R_1$, $R_2$, $R_3$, $R_4$ = H
**b**, $R_1$, $R_2$, $R_3$, $R_4$ = $CH_3$
**c**, $R_1$, $R_3$, $R_4$ = H; $R_2$ = $CH_3$
**d**, $R_1$, $R_3$ = H; $R_2$, $R_4$ = $CH_3$
**e**, $R_1$, $R_2$ = H; $R_3$, $R_4$ = $CH_3$
**f**, $R_1$, $R_2$ = Ac; $R_3$, $R_4$ = $CH_3$

Chromatography of the crude extract of *I. nevadensis* yielded the pseudoguaianolides parthenin (**1**)[8] and coronopilin (**2**),[9,10] the flavone hispidulin (**3c**),[11] and two other flavonoids which were not immediately identifiable. The first of these, obtained in very small amount, was a dihydroxydimethoxyflavone, mp 206–208° (from benzene). The distribution of functional groups on the flavone nucleus as in **3** was suggested by the ultraviolet [$\lambda_{max}$ 277.5 mμ (ε 23,100) and 334 mμ (ε 21,400)][14] and nmr spectrum which exhibited the typical $A_2B_2$ system of H-2′, H-3′, H-5′, and H-6′ at 8.03 and 7.10 ppm and singlets characteristic of H-3 and H-8 at 6.81 and 6.63 ppm[15,16] This was confirmed by conversion to the tetramethyl ether (**3b**) of scutellarein (**3a**).

That one of the two methoxyl groups was attached to C-4′ was indicated by the ultraviolet spectrum in base which did not exhibit the typical bathochromic shift of band I produced by C-4′ hydroxyl.[14] Nmr and infrared spectra (chelated hydroxyl), the brown-green ferric chloride test,[17] and the spectral changes produced by addition of aluminum chloride corroborated the presence of a free hydroxyl group at C-5, which left pectolinarigenin (**3d**) (lit.[18] mp 219°) and **3e** (lit.[19] mp 211–213°) as the remaining possibilities for the flavone isolated from *I. nevadensis*.

However, that the dihydroxydimethoxyflavone from *I. nevadensis* might be identical with pectolinarigenin was considered quite unlikely because of the appreciable difference in melting points and the observation that the effect of sodium acetate and aluminum chloride on the ultraviolet spectrum in ethanol did not produce the changes which the generally accepted rules for structural diagnosis require for pectolinarigenin. Flavones which contain a free 7-hydroxyl group reportedly[14] show a 8–20-mμ bathochromic shift of band II on addition of fused sodium acetate; yet the unknown substance exhibited merely an increase in intensity. Ionization of a 7-hydroxyl group with sodium ethoxide is said[14] to produce a bathochromic shift of 35 mμ in band I and a 30% decrease in intensity as well as a 12-mμ bathochromic shift of band II whose intensity is increased markedly; yet the unknown substance displayed the characteristic changes of band I only. Lastly, addition of aluminum chloride to a flavone containing a 5-hydroxyl group results in considerable bathochromic shifts of both bands with each of the bands showing two distinct peaks or inflections, whereas band II of the unknown substance exhibited two peaks in the presence of aluminum chloride, but band I did not.

(1) Supported in part by a grant from the U. S. Public Health Service (GM-05814). Presented before the Fourth International Symposium on the Chemistry of Natural Products, Stockholm, Sweden, June 1966.

(2) Previous paper, W. Herz, A. Romo de Vivar, and M. V. Lakshmikantham, *J. Org. Chem.*, **30**, 118 (1965).

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(6) This species, previously regarded as the type of the monotypic genus *Oxytenia*, has recently been incorporated into *Iva*.[7]

(7) R. C. Jackson, *Univ. Kansas Sci. Bull.*, **41**, 793 (1960).

(8) W. Herz, H. Watanabe, M. Miyazaki, and Y. Kishida, *J. Am. Chem. Soc.*, **84**, 2601 (1962).

(9) W. Herz and G. Högenauer, *J. Org. Chem.*, **26**, 5011 (1961).

(10) The properties of a third, new sesquiterpene lactone which was isolated in small quantity only are given in the Experimental Section.

(11) Previously isolated from *Ambrosia hispida* Pursh.[12] and *Gaillardia fatigiata* Greene.[13]

(12) W. Herz and Y. Sumi, *J. Org. Chem.*, **29**, 3438 (1964).

(13) W. Herz, S. Rajappa, S. K. Roy, J. J. Schmid, and R. N. Mirrington, *Tetrahedron*, **22**, 1907 (1966).

(14) L. Jurd, "The Chemistry of Flavonoid Compounds," T. A. Geissman, Ed., The Macmillan Co., New York, N. Y., 1962, pp 107–155.

(15) J. Massicot and J. P. Marthe, *Bull. Soc. Chim. France*, 1962 (1962).

(16) T. Batterham and R. J. Highet, *Australian J. Chem.*, **17**, 428 (1964).

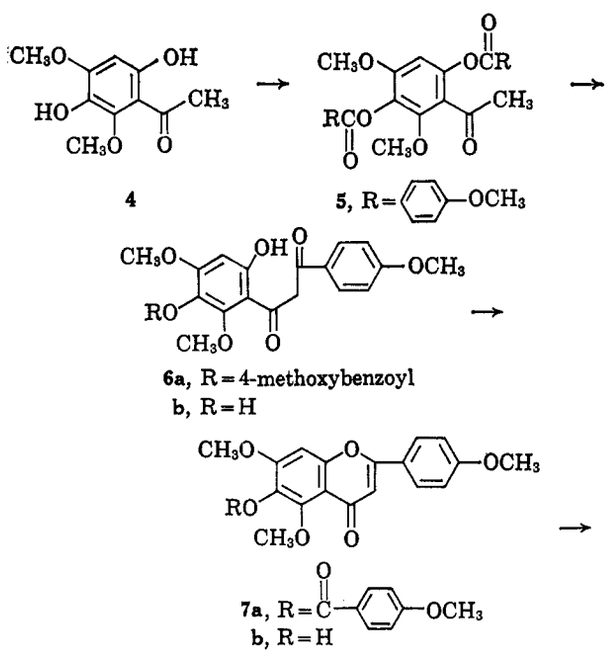
(17) L. H. Briggs and R. H. Locker, *J. Chem. Soc.*, 3136 (1951).

(18) L. Schmid and W. Rumpel, *Monatsh.*, **60**, 8 (1932); V. V. S. Murti and T. R. Seshadri, *Proc. Indian Acad. Sci.*, **30A**, 78 (1949).

(19) V. D. N. Sastri and T. R. Seshadri, *ibid.*, **23A**, 273 (1946).

It appeared therefore that the dihydroxydimethoxyflavone from *I. nevadensis* might be identical with **3e** which had not previously been reported as a natural product. Since the quantity available to us was not sufficiently great to permit degradation, an authentic specimen of **3e** was synthesized for comparison purposes by a new unequivocal route as follows.

2,5-Dihydroxy-4,6-dimethoxyacetophenone (**4**) was acylated with 4-methoxybenzoyl chloride and the resulting 2,5-di(4-methoxybenzoyloxy)-4,6-dimethoxyacetophenone (**5**) was converted to **6a** by the Baker–Venkataraman transformation[20] and thence by ring closure to 6-(4-methoxybenzoyloxy)-4′,5,7-trimethoxyflavone (**7a**). Hydrolysis of **7a** with sodium methoxide furnished 6-hydroxy-4′,5,7-trimethoxyflavone (**7b**) which was partially demethylated with aluminum chloride to **3e**.[21] The melting point of the product coincided with that of the flavone previously synthesized[19] by the Allan–Robinson method, but it was different (mixture melting point, infrared spectrum, tlc) from the dihydroxydimethoxyflavone from *I. nevadensis*.

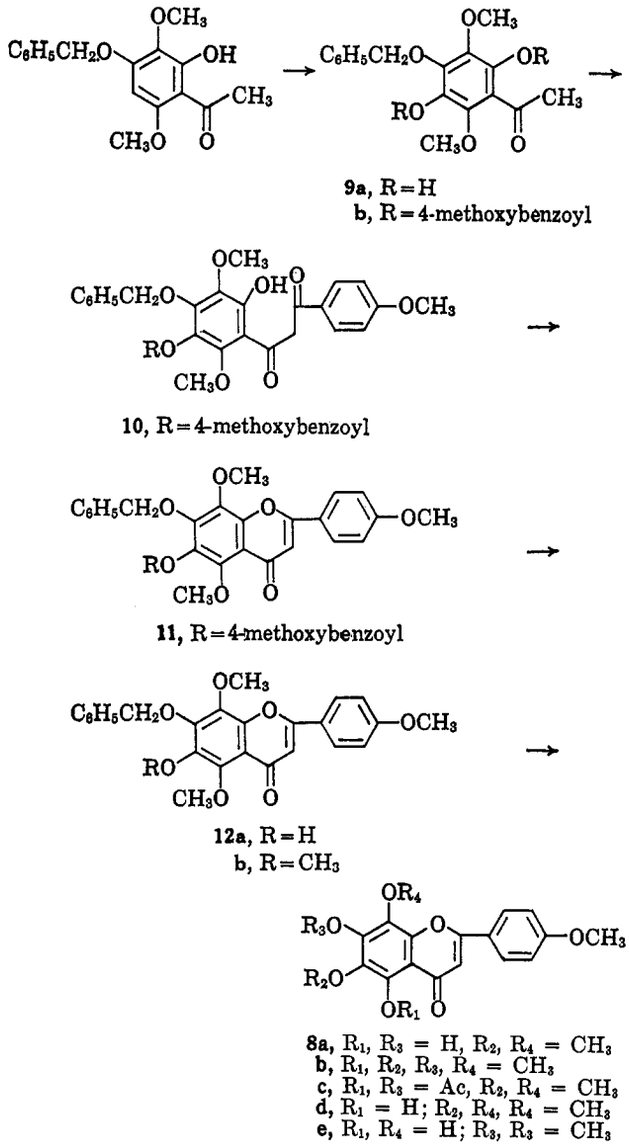



We were therefore forced to doubt the finality of conclusions derived wholly from spectroscopic findings and had to assume that the dihydroxydimethoxyflavone from *I. nevadensis* was pectolinarigenin, in spite of the ultraviolet spectra and the apparent difference in melting point. This supposition was finally confirmed by direct comparison of our material (ultraviolet, infrared, and tlc) with an authentic sample of slightly impure (by tlc criteria) pectolinarigenin, mp 214–217°. Subsequently, it was discovered that the melting point of pure pectolinarigenin when recrystallized from benzene is 206–208°, but that it rises to 217° on recrystallization from methanol, which accounts for the apparent discrepancy in melting point.

The third flavone encountered during chromatography of the crude extract was a previously unknown dihydroxytrimethoxyflavone, mp 193–195°, diacetate

(20) W. Baker, *J. Chem. Soc.*, 1381 (1933); K. Venkataraman and H. S. Mahal, *ibid.*, 1767 (1934).

(21) K. Venkataraman in ref 14, pp 70–106.




mp 170–173°, which we have called nevadensin. The ultraviolet [$\lambda_{max}$ 285 mμ ($\epsilon$ 23,400) and 332 mμ ($\epsilon$ 17,000)] and nmr spectra ($A_2B_2$ quartet at 8.03 and 7.12, singlet at 6.81 ppm, H-3 or H-8) suggested substitution at position 3,4′, 5,6,7, or 4′,5,6,7,8. That the latter was correct was shown by methylation which resulted in tangeretin (**8b**) indistinguishable from an authentic sample.

Spectra and color reactions indicated the presence of a 4′-methoxyl and a 5-hydroxyl group. The first of these points was confirmed by alkaline degradation of nevadensin to anisic acid. A negative gossypetone reaction suggested the presence of a methoxyl group at C-8 as well, and, since the color reaction with *o*-dinitrobenzene was negative, it seemed likely that the third methoxyl group was at C-6 as in **8a**. However, these conclusions were to some extent contraindicated by the same spectral complications (see the Experimental Section) that had dogged the identification of pectolinarigenin, the absence of the band II bathochromic shift in sodium acetate–ethanol being most noteworthy. On the basis of these results one would have been forced to the conclusion that a methoxyl group was attached to C-7.

Since doubt had now been cast on the reliability of spectroscopic criteria, a synthesis of **8a** was therefore

undertaken by the route outlined below in order to clarify the structure of nevadensin. Oxidation of 2-hydroxy-4-benzyloxy-3,6-dimethoxyacetophenone[22] with potassium persulfate led to 2,5-dihydroxy-4-benzyloxy-3,6-dimethoxyacetophenone (**9a**). Acylation of **9a** to **9b** followed by the Baker–Vankataraman transformation[21] led to 4-benzyloxy-2-hydroxy-5-(4-methoxybenzoyloxy)-4′,3,6-trimethoxydibenzoylmethane (**10**) which was not isolated but immediately cyclized to 7-benzyloxy-6-(4-methoxybenzoyloxy)-4′,-5,8-trimethoxyflavone (**11**). Hydrolysis of the latter to **12a** followed by methylation of the hydroxyl group at position 6 afforded 7-benzyloxy-4′,5,6,8-tetramethoxyflavone (**12b**). Debenzylation and demethylation of **12b** at 20° with aluminum chloride in dry ether then furnished 5,7-dihydroxy-4′,6,8-trimethoxyflavone (**8a**) which was identical in all respects with nevadensin.

Methylation of nevadensin with dimethyl sulfate–potassium carbonate furnished 5-hydroxy-4′,6,7,8-tetramethoxyflavone (**8d**). This constitutes an unambiguous total synthesis of another naturally occurring flavone which has recently been isolated from *Citrus jambhiri*.[23]

The unexpected effect of sodium acetate and aluminum chloride on the ultraviolet spectra of pectolinarigenin and nevadensin indicates that caution must be exercised in applying some generally accepted rules for structural diagnosis in the flavone series.

Examination of *I. acerosa* (Nutt). Jackson resulted in the isolation of coronopilin and nevadensin. This appears to offer strong support for the conclusions of Jackson,[7] which were based primarily on morphological evidence, that *I. nevadensis*, *I. acerosa*, and *I. xanthifolia* form a closely related group in the section *Cyclachaena*.[24]

## Experimental Section[25]

**Extraction of *I. Nevadensis* M. E. Jones.**—Ground whole plant, 1.42 kg, from a collection made by Mrs. June McCaskill in September 1964, 50 miles north of Tonopah, Nye County, Nev., was extracted in two portions of 0.74 and 0.68 kg with chloroform for 2 days and worked up in the usual manner.[26] The first portion yielded 22 g of crude gum, the second 50 g.

The gum from portion 1 was taken up in benzene–chloroform and chromatographed over 250 g of silicic acid. The fractions eluted with benzene–chloroform (3:1) solidified on treatment with hexane. Repeated recrystallization from ether–hexane furnished 0.2 g of a sesquiterpene lactone: mp 129–131°; $[\alpha]^{27}D$ +173.5° (c 1.27, $CHCl_3$); $\lambda_{max}$ 292 mμ (ε 39); infrared bands at 1770 (γ-lactone), 1745 (cyclopentanone), and 1410 $cm^{-1}$ ($C(O)CH_2$); nmr signals at 4.6 c (H α to lactone oxygen), 1.23 d and 1.11 d (methyl doublets, $J$ = 7 cps), and 1.00 ppm (methyl singlet). The substance gave a positive Zimmerman test and differed from other compounds previously isolated in our laboratory. The quantity available was not sufficient to permit structural elucidation, but the nmr spectrum suggested the presence of a pseudoguaianolide skeleton containing a ketone in the five-membered ring and lactone ring closure to $C_8$ because of the multiplicity of the signal at 4.6 ppm.

*Anal.* Calcd for $C_{15}H_{22}O_3$: C, 71.97; H, 8.86; O, 19.17. Found: C, 72.41; H, 8.90; O, 19.10.

The fractions eluted with benzene–chloroform (5:2) solidified on trituration with ether and were recrystallized repeatedly from benzene. This furnished 0.2 g of nevadensin which exhibited a double melting point of 186–188° and 193–195° (block); infrared bands at 1655 and 1590 $cm^{-1}$; $\lambda_{max}$ 285 mμ (ε 23,400) and 332 mμ (ε 17000); $\lambda_{max}$ (with fused sodium acetate) 285 mμ (ε 30,500), 300 (24,100) (inflection), and 382 (12500); $\lambda_{max}$ (with sodium hydroxide) 284 mμ (ε 28,200), 300 (inflection) (25,500), and 380 (12700); $\lambda_{max}$ (with $AlCl_3$) 235 mμ (ε 13,400), 268 (inflection) (8400), 313 (11,300), and 361 (11,200); nmr bands (in deuteriodimethyl sulfoxide) at 12.72 ($C_5$-OH), 8.03 d and 7.12 d ($J$ = 9 cps, AB quartet, H-2′, H-3′, H-5′, and H-6′), 6.81 s (H-3), 3.93, 3.88, and 3.85 ppm (three methoxyl singlets). The substance gave the following color reactions: sodium hydroxide, intense yellow; magnesium and hydrochloric acid, yellowish red; ferric chloride, green with blue fluorescence; gossypetone reaction and reaction with *o*-dinitrobenzene and aqueous sodium carbonate, negative.

*Anal.* Calcd for $C_{18}H_{16}O_7$: C, 62.79; H, 4.68; O, 32.53. Found: C, 63.19; H, 4.59; O, 32.32.

The fractions eluted with benzene–chloroform (1:1) solidified on trituration with ether. Repeated recrystallization from chloroform–hexane furnished 0.4 g of coronopilin, mp 175–178°, identical in infrared and nmr spectra and in mixture melting point with authentic material. Chloroform–benzene (2:1) eluted gummy material which on recrystallization from chloroform–ether furnished 0.25 g of parthenin, mp 165°, indistinguishable by mixture melting point, infrared, and nmr spectroscopy from authentic parthenin.

The gum from portion 2 was chromatographed over silicic acid in the same manner, but all fractions remained mixtures (tlc) in which because of many overlapping spots it was not possible to establish the presence of the substances isolated from the first chromatogram. We ascribe this to minor differences in the work-up which apparently resulted in the isolation of a more complex mixture of constituents. Rechromatography of the fractions eluted with benzene, benzene–chloroform (3:1), and benzene–chloroform (2:1) over silicic acid gave in the benzene–ether (5:1) eluate a gummy mixture (tlc) which dissolved in petroleum ether (bp 30–60°) containing a small amount of ether. Leaving this at room temperature for some weeks resulted in the separation of a small amount of pectolinarigenin which was recrystallized several times from benzene: yield 23 mg; mp 206–208° (only one spot on tlc), mp 217° after recrystallization from methanol (lit.[18,19] mp 219°); nmr bands (in deuteriodimethyl sulfoxide) at 8.03 d and 7.10 d ($J$ = 9 cps, $A_2B_2$ quartet, H-2′, H-3′, H-5′, and H-6′), 6.81 s (H-3), 6.63 (H-8), 3.87, and 3.80 ppm (two methoxyl singlets); mixture melting point with a slightly impure sample (tlc) of authentic pectolinarigenin (mp 214–217°), 212–216°; infrared, nmr, and ultraviolet spectra identical.

*Anal.* Calcd for $C_{17}H_{14}O_6$: C, 64.98; H, 4.68; O, 30.38. Found: C, 64.96; H, 4.49; O, 30.55.

Methylation of 11 mg of pectolinarigenin by refluxing in 50 ml of acetone with 2 g of anhydrous potassium carbonate and 1 ml of methyl iodide for 50 hr, filtration, evaporation, and extraction of the residue with water and then with ether gave, from the ether extract, an oil which was chromatographed over 5 g of silicic acid. Benzene–chloroform (3:1) eluted material which was recrystallized from acetone–ether–hexane: mp 160–162°; identical in melting point, mixture melting point, and infrared spectrum with tetramethylscutellarein; nmr bands at 7.82 d and 7.00 ($J$ = 8.5 cps, $A_2B_2$ quartet, H-2′, H-3′, H-5′, and H-6′), 6.81 s (H-3), 6.60 s (H-8), 3.99 s (two methoxyls), 3.92, and 3.89 ppm (two methoxyl singlets).

The fractions eluted from the chromatogram of portion 2 with chloroform–methanol (195:5) gave a yellow solid which was recrystallized from methanol to yield 10 mg: mp 288–290°; identified by mixture melting point and infrared and nmr spectra as hispidulin; nmr signals (in deuteriodimethyl sulfoxide) at 7.95 d and 6.95 d ($J$ = 8.5 cps H-2′, H-3′, H-5′, and H-6′), 6.77 s (H-3), 6.62 s (H-8), and 3.8 ppm (methoxyl singlet).

**Characterization of Nevadensin.**—A mixture of 60 mg of nevadensin, 80 ml of dry acetone, 3 g of anhydrous potassium carbonate, and 2 ml of methyl iodide was refluxed for 36 hr. Fresh amounts of potassium carbonate (0.5 g) and methyl iodide (0.5 ml) were added and refluxing was continued for another 24 hr. The mixture was filtered, the residual solid was

(22) N. Rabjohn and D. W. Rosenberg, *J. Org. Chem.*, **24**, 1192 (1959).

(23) B. P. Chaliha, G. P. Sastri, and P. R. Rao, *Tetrahedron*, **21**, 1441 (1965).

(24) Our previous studies of *Iva* species belonging to the sections *Linnearbractea* and *Iva* have led only to the isolation of eudesmanolides and guaianolides. On the other hand an investigation of *I. xanthifolia* Nutt. (W. Herz, unpublished) which like *I. nevadensis* and *I. acerosa* belongs to the section *Cychlachaena* led to the isolation of coronopilin.

(25) Melting points are uncorrected. Ultraviolet spectra were determined in absolute ethanol; infrared spectra were determined in chloroform unless otherwise specified. All isolations and reactions were controlled by thin layer chromatography (Merck silica gel G, benzene–ethyl acetate, 4:1).

(26) W. Herz and G. Högenauer, *J. Org. Chem.*, **27**, 905 (1965).

washed with acetone, and the combined filtrate and washings were evaporated. The residue was taken up in water and extracted with ether, and the dried ether extract was evaporated. The residue was recrystallized from acetone–hexane to yield 30 mg: mp 152–154° (lit.[27] mp 152–153.5°); nmr signals at 7.91 d and 7.06 d ($J$ = 9 cps, $A_2B_2$ quartet, H-2′, H-3′, H-5′, and H-6′), 6.61 s (H-3), 4.12 (3p), 4.04 (3p), 3.96 (6p), and 3.88 ppm (3p, five methoxyls); infrared spectrum identical with that of authentic samples of tangeretin;[28] mixture melting point undepressed.

A mixture of 30 mg of nevadensin, 3 ml of acetic anhydride, and 1 g of sodium acetate was refluxed for 3 hr, cooled, and poured on ice. The product was filtered, washed with water, dried, and chromatographed over 5 g of silicic acid. The residue obtained by evaporation of the benzene–ether (5:1) eluates was recrystallized from acetone–ether–hexane: yield 20 mg; mp 170–173°; infrared bands at 1760 (acetate, strong), 1645, and 1602 $cm^{-1}$; nmr signals at 7.92 d and 7.06 d ($J$ = 9 cps, $A_2B_2$ quartet), 6.62 s (H-3), 4.06 s (methoxyl), 3.88 s (6p, two methoxyls), 2.5 s (3p), and 2.45 ppm (3p, two acetates).

*Anal.* Calcd for $C_{22}H_{20}O_9$: C, 61.68; H, 4.71; O, 33.61. Found: C, 61.57; H, 5.04; O, 33.72.

A mixture of 30 mg of nevadensin and 10 ml of 50% aqueous sodium hydroxide was refluxed for 4 hr in a nitrogen atmosphere, cooled, acidified, concentrated to small volume, and extracted with chloroform. The extract was evaporated and the residue was dissolved in sodium bicarbonate solution and extracted with chloroform. The aqueous layer was acidified and extracted with chloroform, and the dried chloroform extract was evaporated. The residue was chromatographed over 5 g of silicic acid and the material was isolated from the benzene–ether (4:1) eluate recrystallized from acetone–hexane. The product (6 mg) melted at 184–186°; melting point undepressed on admixture of anisic acid; infrared spectra superimposable.

**2,5-Di(4-methoxybenzoyloxy)-4,6-dimethoxybenzoyloxyacetophenone (5).**—A mixture of 2.12 g of 2,5-dihydroxy-4,6-dimethoxyacetophenone[29] and 3.7 g of 4-methoxybenzoyl chloride was heated in 6.5 ml of dry pyridine for 10 min in an oil bath at 110°. The cooled mixture was diluted with 15 ml of methanol, whereupon the product crystallized as colorless prisms, mp 210–211°, from chloroform–methanol, yield 3.4 g (72%).

*Anal.* Calcd for $C_{26}H_{24}O_9$: C, 64.99; H, 5.04. Found: C, 65.10; H, 5.05.

**2-Hydroxy-5-(4-methoxybenzoyloxy)-4′,4,6-trimethoxydibenzoylmethane (6a).**—A mixture of 4.8 g of 5 and 0.77 g of powdered potassium hydroxide was heated for 3 hr at 60° with stirring. The brownish solution was poured into 140 ml of 20% aqueous acetic acid and the yellow resin which separated dissolved in chloroform. The chloroform solution was dried and evaporated and the residue recrystallized from acetic acid. The product formed yellowish green prisms: mp 141–143°; ferric chloride reaction (in methanol) green; $\lambda_{max}$ 264 mμ (log ε 4.35) and 392 mμ (log ε 4.36).

*Anal.* Calcd for $C_{26}H_{24}O_9$: C, 64.99; H, 5.04. Found: C, 64.80; H, 5.10.

**2,5-Dihydroxy-4′,4,6-trimethoxydibenzoylmethane (6b).**—A mixture of 0.1 g of 6a and 6 ml of 0.1 $N$ sodium methoxide solution was refluxed for 30 min. After acidification with acetic acid, the product gradually crystallized in fine, yellow needles. Recrystallization from dimethylformamide–methanol furnished material: mp 275–277°, with previous sublimation at 240° (microhot stage); brown ferric chloride reaction in methanol solution; $\lambda_{max}$ 223 mμ (log ε 4.24), 283 (4.26), and 348 (3.82, inflection).

*Anal.* Calcd for $C_{18}H_{18}O_7$: C, 62.42; H, 5.24. Found: C, 62.70; H, 5.20.

**6-(4-Methoxybenzoyloxy)-4′,5,7-trimethoxyflavone (7a).**—A solution of 0.48 g of 6a in 6 ml of ethanol containing 0.2 ml of sulfuric acid was refluxed for 1 hr. Precipitation of the product began before refluxing had been completed; yield of almost pure flavone was 0.40 g (87%). Recrystallization from acetic acid and subsequently from methanol–chloroform afforded small colorless prisms, mp 234–236°.

*Anal.* Calcd for $C_{26}H_{22}O_8$: C, 67.52; H, 4.80. Found: C, 67.27; H, 4.69.

**6-Hydroxy-4′,5,7-trimethoxyflavone (7b).**—A solution of 1.2 g of 7a in 10 ml of 1 $N$ sodium hydroxide solution was refluxed for 30 min. On acidification with acetic acid the free flavone precipitated immediately and was recrystallized from ethanol, yielding 0.80 g (94%), mp 218–220° (lit.[30] mp 217–218°).

**5,6-Dihydroxy-4′,7-dimethoxyflavone (3e, Scutellarein 4′,7-Dimethyl Ether).**—A solution of 0.2 g of 7b and 0.1 g of anhydrous aluminum chloride in 2 ml of nitrobenzene was heated at 105° for 1 hr. The solution was added to 10 ml of dilute hydrochloric acid and the solvent was removed by steam distillation. The crystalline product was recrystallized from a small amount of acetic acid as bright yellow, long, flat, rectangular prisms: mp 211.5–213° (lit.[19] mp 211–213°); green ferric chloride reaction in methanol; $\lambda_{max}$ 285 mμ (log ε 4.40) and 330 mμ (log ε 4.43). The crude product was homogeneous on tlc, indicating selective demethylation of C-5 methoxyl.

**5,6-Diacetoxy-4′,7-dimethoxyflavone (3f).**—Acetylation of 30 mg of 3e in the usual way with acetic anhydride–sodium acetate furnished colorless needles from ethanol, mp 210–212°.

*Anal.* Calcd for $C_{21}H_{28}O_8$: C, 63.31; H, 4.55. Found: C, 63.50; H, 4.50.

**2-Hydroxy-4-benzyloxy-3,6-dimethoxyacetophenone.**—A mixture of 2,4-dihydroxy-3,6-dimethoxyacetophenone,[31] 60 ml of dimethylformamide, 6.6 ml of benzyl chloride, and 12 g of anhydrous potassium carbonate was refluxed for 1 hr with stirring and poured into 600 ml of water. The precipitate was recrystallized from 100 ml of ethanol, yield 12.1 g (70%), mp 110–112° (lit.[22] mp 111–111.5°).

**2,5-Dihydroxy-4-benzyloxy-3,6-dimethoxyacetophenone (9a).**—To a suspension of 4.0 g of the previous compound in 40 ml of water containing 2.64 g of sodium hydroxide there was added a solution of 3.9 g of potassium persulfate in 80 ml of water at 5–10° over a 4-hr period with constant stirring. After stirring had been continued for another 30 min, the mixture was acidified with concentrated hydrochloric acid to pH 4 and the precipitate (unchanged starting material, 2.2 g) was removed by filtration. The aqueous solution was extracted with chloroform (two 10-ml portions) to remove benzaldehyde and other by-products. Finally 1 g of sodium sulfite, 22 ml of concentrated hydrochloric acid, and 40 ml of chloroform was added and the mixture was refluxed for 1 hr. The chloroform layer was separated, extracted with 5% sodium carbonate solution, and dried. Evaporation of solvent yielded oily material (0.6 g), which was almost pure as shown by tlc. Addition of petroleum ether effected slow crystallization. The yellow needles melted at 60–62°.

*Anal.* Calcd for $C_{17}H_{18}O_6$: C, 64.14; H, 5.70. Found: C, 63.90; H, 5.65.

**4-Benzyloxy-3,6-dimethoxy-2,5-di(4-methoxybenzoyloxy)acetophenone (9b).**—A mixture of 0.8 g of 9a and 1.5 g of anisoyl chloride in 3 ml of dry pyridine was heated on the water bath for 30 min and poured onto 20 ml of 5% hydrochloric acid. The precipitate was taken up in chloroform, the chloroform solution was evaporated, and the residue was extracted with hot methanol (two 10-ml portions). The product (1.0 g, 64%) crystallized from the methanol solution and was sufficiently pure for further work. Recrystallization of a small sample from methanol–acetone afforded colorless prisms, mp 132–134°.

*Anal.* Calcd for $C_{33}H_{30}O_{10}$: C, 67.57; H, 5.16. Found: C, 67.23; H, 5.18.

**7-Benzyloxy-6-(4-methoxybenzoyloxy)-4′,5,8-trimethoxyflavone (11).**—A mixture of 0.6 g of 9b and 78 mg of powdered potassium hydroxide was heated with 3 ml of pyridine for 3 hr at 60° with stirring and poured into 10 ml of 2.5% hydrochloric acid. The crude yellow dibenzoylmethane 10 which separated was refluxed for 1 hr with 6 ml of 1.5% ethanolic sulfuric acid. Crystallization of the product began at the very beginning of the reflux period; the yield was 0.55 g (93%). Recrystallization of a small sample from ethanol–chloroform yielded colorless rhomboidal prisms, mp 188–189°.

*Anal.* Calcd for $C_{33}H_{28}O_9$: C, 69.71; H, 4.96. Found: C, 69.85; H, 5.10.

**6-Hydroxy-7-benzyloxy-4′,5,8-trimethoxyflavone (12a).**—A solution of 0.5 g of 13 in 3 ml of 1 $N$ sodium methoxide was refluxed for 1 hr. Acidification of the cooled solution with acetic acid immediately precipitated the free flavone as long, colorless

(27) J. Gripenberg in ref 14, p 426.

(28) Kindly supplied by Dr. L. Swift, U. S. Fruit and Vegetable Laboratory, Winter Haven, Fla., and Dr. R. M. Horowitz, U. S. Fruit and Vegetable Chemistry Laboratory, Pasadena, Calif.

(29) V. D. N. Sastri and T. R. Seshadri, *Proc. Indian Acad. Sci.*, **23A**, 262 (1946).

(30) M. G. Stout, H. Reich, and M. N. Huffman, *J. Pharm. Sci.*, **53**, 192 (1964).

(31) V. D. N. Sastri and T. R. Seshadri, *Proc. Indian Acad. Sci.*, **24A**, 248 (1946).

needles. The product was filtered and washed with cold methanol until the characteristic odor of methyl anisate could no longer be detected, yield 0.37 g (97%). The analytical sample was recrystallized from ethanol, mp 172–173°.

*Anal.* Calcd for $C_{25}H_{22}O_7$: C, 69.11; H, 5.10. Found C, 68.92; H, 5.05.

**7-Benzyloxy-4′,5,6,8-tetramethoxyflavone (12b).**—A mixture of 0.34 g of **12a**, 50 ml of acetone, 2 ml of dimethyl sulfate, and 2 g of anhydrous potassium carbonate was refluxed for 3 hr with stirring. The inorganic salts were filtered, the solvent was removed, and the residue was chromatographed over 25 g of silicic acid (solvent benzene–ethyl acetate, 4:1) in order to remove a green contaminant. Evaporation of the eluate afforded 0.25 g (70%) of the product. Recrystallization from benzene gave small, elongated prisms, mp 114–116°.

*Anal.* Calcd for $C_{26}H_{24}O_7$ C, 69.63; H, 5.39. Found: C, 69.14; H, 4.90.

**5,7-Dihydroxy-4′,6,8-trimethoxyflavone (8a, Nevadensin).**—On addition of 0.23 g of **12b** to a cold solution of 4 g of anhydrous aluminum chloride in 20 ml of dry ether, the ether-insoluble flavone dissolved gradually. After 3 hr the solvent was removed and the residue was treated with 50 ml of ice cold dilute hydrochloric acid (1:1). After heating for about 5 min on the water bath, the precipitate was filtered, dried, and chromatographed over 25 g of silicic acid (solvent benzene–ethyl acetate, 4:1). Evaporation of the less polar fraction yielded 95 mg of the almost pure product, mp 193–195°. Recrystallization from benzene raised the melting point to 197–198° (Kofler block); $\lambda_{max}$ 284 mμ (log ε 4.38 and 334 mμ log ε 4.24). The mixture melting point with authentic nevadensin was undepressed and the infrared spectra were superimposable.

**5,7-Diacetoxy-4′,6,8-trimethoxyflavone (8c, Nevadensin Diacetate).**—The totally synthetic material was prepared in the manner previously described for the preparation of the derivative of nevadensin, mp 171–174°, mixture melting point undepressed.

**5-Hydroxy-4′,6,7,8-tetramethoxyflavone (8d, 5-Desmethyltangeretin).**—A solution of 22 mg of nevadensin in 5 ml of acetone containing 30 mg of anhydrous potassium carbonate and 8.5 mg of dimethyl sulfate was stirred under reflux for 1 hr. The inorganic salts were filtered, the solvent was evaporated, and the residue was recrystallized from ethanol. The product long yellow needles: mp 175–177° (lit.[32] mp 174–175°), $\lambda_{max}$ 292 mμ (log ε 4.38) and 328 mμ (log ε 4.33).

**Extraction of *I. Acerosa* (Nutt.) Jackson.**—Above-ground material, collected in 1964 by members of the Agricultural Research Service of Utah State University in the vicinity of Logan, Utah, was made available through the courtesy of Professor F. R. Stermitz. Extraction of 950 g of ground plant in the usual manner[26] furnished 30.5 g of gum which was chromatographed over 225 g of silicic acid. Fractions 1–5 (400 ml of benzene each) yielded nothing, fractions 6–12 (benzene) yielded 0.2 g of what appeared to be triterpene mixture (positive Noller test), fractions 13–21 (benzene–chloroform, 3:1) yielded traces only, and fractions 22–26 (benzene–chloroform, 2:1) gave a residue which solidified on trituration with ether. Recrystallization from benzene yielded nevadensin, mp 186–188° and 193–195°, yield 0.15 g, mixture melting point undepressed, infrared and nmr spectra superimposable. Fractions 27–31 (benzene–chloroform, 2:1) gave gum; fractions 32–37 (benzene–chloroform, 1:1) gave a residue which solidified on trituration with ether. Recrystallization from acetone–ether–hexane afforded coronopilin, yield 0.3 g, mp 174–176°, mixture melting point undepressed, infrared and nmr spectra superimposable.

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(32) Chaliha, *et al.*,[23] prepared this flavone from *Citrus Jambhiri* and prepared it from tangeretin by demethylation with aluminum chloride and from 5,8-dihydroxy-4′,6,7-trimethoxyflavone (**8e**) by methylation with diazomethane. Compound **8e** in turn was prepared by nitric acid oxidation of tangeretin by reduction with sodium bisulfite. A previous preparation[33] of **8e** involved persulfate oxidation of 5-hydroxy-4′,6,7-trimethoxyflavone.

(33) V. V. S. Murti, K. V. Rao, and T. R. Seshadri, *Proc. Indian Acad. Sci.*, **26A**, 182 (1947).

# Constituents of *Iva* Species. VII. New Guaianolides from *Iva axillaris* Pursh. ssp. *robustior*[1,2]

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The structure of three new guaianolides which were isolated from *Iva axillaris* Pursh. ssp. *robustior* has been established.

In earlier parts of our systematic study of the genus *Iva*, we reported the isolation of eudesmanolides from *Iva microcephala* Nutt.,[3,4] *I. imbricata* Walt.,[3] *I. asperifolia* Less.,[5] and *I. texensis* Jackson,[5] the isolation of guaianolides from a variety of *I. microcephala*,[6] and the isolation of pseudoguaianolides from *I. acerosa* (Nutt.) Jackson, *I. nevadensis* M. E. Jones, and *I. xanthifolia* Nutt.[7] We now describe the isolation and structure determination of new guaianolides from *I. axillaris* Pursh. ssp. *robustior*.[8]

Extraction of *I. axillaris* ssp. *robustior* collected in Reno, Nev., furnished three new guaianolides, which we have called axivalin, ivaxillin, and ivaxillarin, and the eudesmanolide microcephalin (**1**).[6] Material collected at Montgomery Pass, Mineral Co., Nev., yielded axivalin, ivaxillin, ivaxillarin, a fourth guaianolide anhydroivaxillarin, and a new flavone axillarin (**2**) which we have described elsewhere.[2] Structures of axivalin, ivaxillarin, and anhydroivaxillarin have been determined and are reported in this paper.

The physical properties of the most plentiful and relatively polar material ivaxillarin (**3**), $C_{15}H_{18}O_4$, mp 186–188°, $[\alpha]^{27}D$ −240.8° (*c* 0.72, $CH_3OH$), high intensity ultraviolet absorption at 207 mμ (ε 10,050), and infrared bands at 1775 and 1665 $cm^{-1}$, suggested

(1) Supported in part by a grant from the U. S. Public Health Service (GM-05814).

(2) Previous paper: W. Herz, L. Farkas, V. Sudarsanam, H. Wagner, L. Hörhammer, and R. Rüger, *Chem. Ber.*, in press.

(3) W. Herz and G. Högenauer, *J. Org. Chem.*, **27**, 905 (1962).

(4) W. Herz, G. Högenauer, and A. Romo de Vivar, *ibid.*, **29**, 1700 (1964).

(5) W. Herz and N. Viswanathan, *ibid.*, **29**, 1012 (1964).

(6) W. Herz, A. Romo de Vivar, and M. V. Lakshmikantham, *ibid.*, **30**, 118 (1965).

(7) L. Farkas, M. Nogradi, V. Sudarsanam, and W. Herz, *ibid.*, **31**, 3228 (1966).

(8) Evidence for the existence of two subspecies, ssp. *robustior* and ssp. *axillaris*, has been presented recently.[9] Our material came from western Nevada where ssp. *axillaris* does not occur.

(9) I. J. Bassett, G. A. Mulligan, and C. Frankton, *Can. J. Botany*, **40**, 1243 (1962).